



# CEREAL CHEMISTRY

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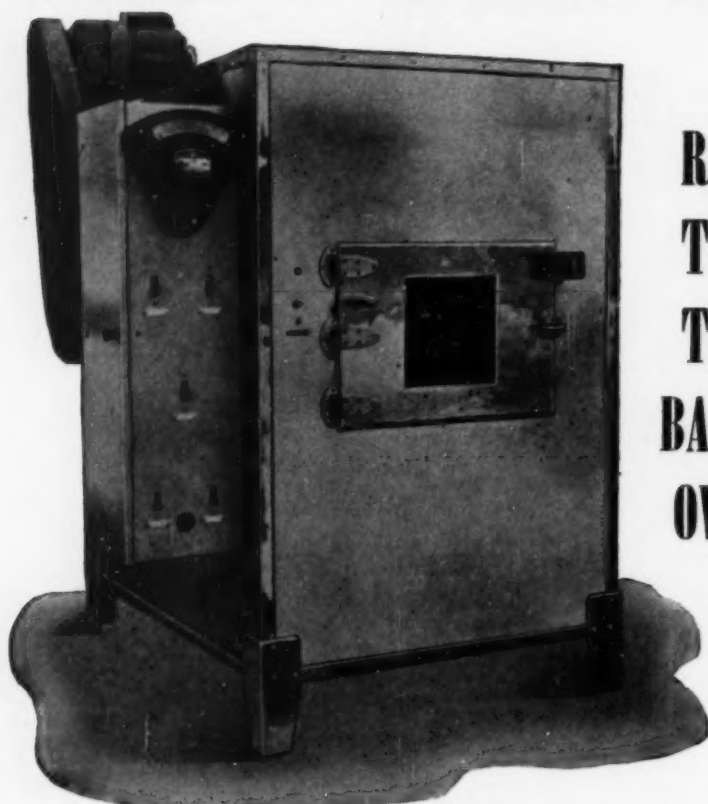
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Vol. 31

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No. 4

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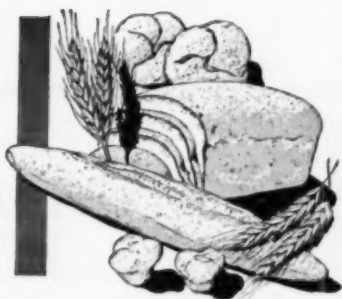
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There came a time when man decided not to migrate in search of new hunting grounds when his game supply became scarce.

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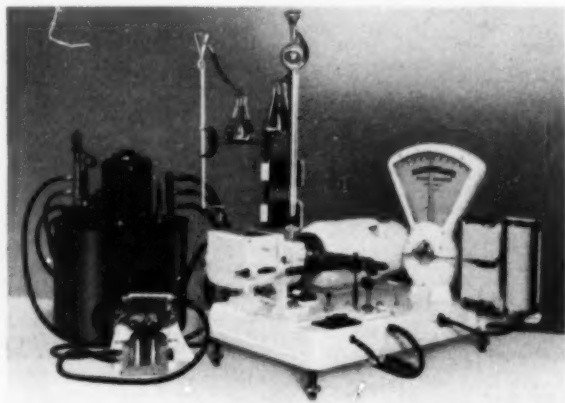
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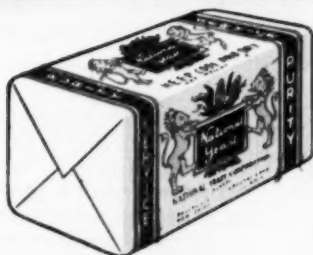


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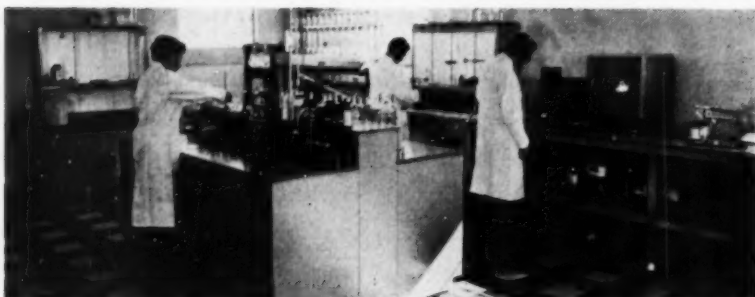


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# CEREAL CHEMISTRY

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## THE BROWNING REACTION IN WHEAT GERM IN RELATION TO "SICK" WHEAT<sup>1</sup>

C. E. McDONALD AND MAX MILNER<sup>2</sup>

### ABSTRACT

The effect of temperature, moisture content, and mold growth on the formation of the brown discoloration in wheat germ which characterizes commercial sick wheat was investigated by means of controlled storage and respiration technics. Extraction of germ with water inhibited the typical discoloration, whereas extraction with ether did not. At constant temperatures of 30°, 40°, and 52°C., and at moisture contents varying from 14 to 20%, the onset of browning in fresh unprocessed wheat germ was promoted by elevated temperature and moisture content, and invariably preceded mold growth. The appearance of the latter was detected by its characteristic acceleration of the respiratory rate.

Progressive browning in wheat germ was characterized by increases in fluorescence and absorbance (245m $\mu$ ) of acid extracts, by a fall in pH, and by a decrease in protein peptizable by potassium sulfate solution. Molds growing in damp stored wheat germ caused respiratory quotient values to approach unity, whereas browning was accompanied by values of 0.5 or less. In the presence of sodium bisulfite, browning and the associated increases in absorbance and fluorescence were inhibited but the characteristic decrease in protein solubility was unaffected, indicating that at least two stages in the browning reaction exist. Freshly wetted germ exhibited a high initial rate of carbon dioxide production, characterized by very high R. Q. values, which is strongly inhibited by bisulfite.

It is concluded that sugar-protein condensation in the germ of stored wheat, favored by elevated moisture and temperature levels, is the basic cause of the brown discoloration of sick wheat. Under practical conditions, the respiration of storage fungi may play a role in promoting this deterioration by providing the elevated temperatures and humidities favorable to the nonbiological browning mechanism.

Wheat which has deteriorated in storage, of the type known in the grain trade as "sick," is recognized by the turning in color of the germ from normal light yellow to light tan, brown, and finally to a dark mahogany. This damage reduces the commercial grade of the grain

<sup>1</sup> Manuscript received December 28, 1953. Contribution No. 245, Department of Flour and Feed Milling Industries, Kansas Agricultural Experiment Station, Manhattan. Presented at the Annual Meeting of the Institute of Food Technologists, Boston, Mass., June 1953. This research is a portion of a thesis presented by Clarence E. McDonald as partial fulfillment of the requirement for the Degree of Master of Science in Milling Industries at Kansas State College.

<sup>2</sup> Respectively, Research Assistant and Professor of Milling Industries, Kansas State College, Manhattan.

and is most prevalent in years when damp harvest conditions are encountered.

Previous investigations have shown mold growth to be associated with this deterioration. Thomas (18) believed "sick" wheat was grain which had lost its viability through toxic products secreted by molds. Milner *et al.* (9) found *Aspergillus glaucus* and *A. flavus* to be the most abundant molds in "sick" wheat kernels, whereas *Alternaria* was most prevalent in sound wheat. Some of the literature indicates, however, that "sick" wheat can develop under atmospheres where molds grow with difficulty (3) and when mold growth is inhibited by chemical preservatives (8, 17).

Cole and Milner (4) recently showed that the absorbance and fluorescence properties of aqueous extracts of "sick" wheat kernels and those of wheat germ browned in storage appear to be identical and that these manifestations are due to a nonenzymatic browning reaction of the Maillard type involving condensation of reducing sugars and proteins in the intact germ. Browning of this kind occurs in many other foodstuffs. A comprehensive review of mechanisms of browning in foods and in model systems has recently been published (6). The present study was undertaken to extend the work of Cole and Milner concerning the color change in germ associated with sick wheat and, by means of respiration technics using fresh granular wheat germ, to clarify the relationship of mold growth to the characteristic browning.

### Materials and Methods

Fresh granular wheat germ used in these studies was obtained from General Mills, Inc., Minneapolis, Minnesota.

To study the relationship of mold growth to the browning effect, storage studies were conducted in respirometers in which temperature, humidity, and aeration rate could be closely controlled. Apparatus similar to that described by Milner and Geddes (10) was used. The air collected after passage through each germ sample was analyzed daily for carbon dioxide and oxygen by a Haldane technic as described by these authors. Mold growth was detected by its characteristic effect on the trend of the respiration curve. Respiration is considered to be a reliable means for detecting a growing biological population (16).

Tempering of germ samples to desired moisture values was accomplished by whirling the material with a stirrer in an Erlenmeyer flask while moistening it with a spray of water from an atomizer. Moisture was determined by the 130°C. air oven method (1).

The fluorescence and absorbance at  $245\text{ m}\mu$  of aqueous extracts of the wheat germ samples were determined by the method outlined by Cole and Milner (4). Certain changes in these optical properties are generally considered to characterize browning in natural systems (14, 15). A method for assaying the peptizable protein content of wheat germ was developed during the course of this study, based on a turbidity method for protein content of flour proposed by Zeleny (19). Such an analysis appeared of interest in view of the known involvement of proteins in the browning phenomenon. A 0.70-g. sample of wheat germ which had been ground to pass a No. 30 screen (Wiley intermediate mill) was shaken intermittently for 15 minutes with 50 ml. of a 5% potassium sulfate solution. The mixture was filtered through No. 4 Whatman filter paper, and 3 ml. of the filtrate were pipetted into a Coleman spectrophotometer tube containing 10 ml. of hydrochloric acid-sodium citrate buffer (pH 2.4). The resulting turbid solution was allowed to stand for 20 to 45 minutes. The tubes were then fitted with rubber stoppers and the contents mixed by three to four inversions of each tube. The light absorption read on a Coleman spectrophotometer at a wave length of  $530\text{ m}\mu$  was directly proportional to the amount of protein peptized.

To prepare the germ for pH determination, 0.5 g. was ground in a mortar with 5 ml. of distilled water. After the wheat germ slurry had stood for 45 to 60 minutes its pH was determined with a Beckman Model G pH meter.

### Results

*Influence of Extraction on Color Changes and Other Properties of Stored Wheat Germ.* Wheat germ contains abundant quantities of soluble sugars (principally sucrose and raffinose), fat, and protein. To determine which of these major constituents might be involved in the darkening of wheat germ, a preliminary storage experiment was carried out with separate portions of ground germ samples extracted repeatedly before storage with cool petroleum ether and with water. Presumably most of the oils or sugars were removed by these respective treatments. Samples of ether-extracted, water-extracted, and ether-water extracted germ samples were stored in constant-humidity jars at 60 and 90% humidities and at  $42^\circ$  and  $10^\circ\text{C}$ .

All the samples stored at 60% humidity changed very little in color during 8 months of storage. Minor discoloration was evident in samples stored at 90% humidity at  $10^\circ\text{C}$ . After 7 months' storage at  $42^\circ\text{C}$ . and 90% humidity, the water-extracted sample had changed to a salmon color, the ether-extracted sample to a dark mahogany brown,

and the water-ether extracted sample to a light brown.

The fluorescence of extracts of these samples are shown in Table I. An extremely high fluorescence value (1170) was obtained with the ether-extracted sample stored at 90% humidity and 42°C. which had turned to dark mahogany brown. Much lower values were obtained with the samples extracted with water which showed a salmon rather than brown discoloration. All samples stored at 60% humidity changed very little during 8 months of storage. These preliminary results suggested that removal of soluble sugars and possibly some of the protein from the germ prior to storage prevents the development of the typical browning and fluorescence of sick wheat, whereas removal of fats apparently has no effect on the color of fluorescence.

TABLE I  
INFLUENCE OF EXTRACTION ON DEVELOPMENT OF FLUORESCENCE  
IN STORED WHEAT GERM

Storage Conditions	Method of Extraction		
	Ether	Water	Ether and Water
60% R.H., 8 mos., 10°C.	3.1	3.3	4.5
60% R.H., 8 mos., 42°C.	16.6	11.2	10.4
90% R.H., 7 mos., 10°C.	30.7	12.4	4.3
90% R.H., 7 mos., 42°C.	1170.0	27.1	36.7

*Storage of Wheat Germ in Respirometers.* To investigate the relationship of browning to mold growth at various temperatures, duplicate pairs of germ samples at three moisture values (approximately 14%, 16%, and 20%) were prepared and stored with aeration in the respirometers at 30°, 40°, and 52°C. for periods as long as 21 days. In the course of these experiments, aliquots from one sample of each moisture level were removed at various time intervals for fluorescence, colorimetric, pH, and peptizable protein measurements. Gas exchange data were obtained daily on the three undisturbed samples. Although certain significant differences in results were obtained at the different storage temperatures, as will be indicated, the data for the trial at 40°C. are broadly representative of these experiments and will therefore be presented in detail.

The carbon dioxide evolution at 40°C., as shown in Fig. 1, is initially high and similar to that previously noted with freshly wetted peas (5) and soybeans (11). A correspondingly strong oxygen uptake occurred which was of lower magnitude than the carbon dioxide evolution. The respiratory quotient values as shown in Fig. 2 were initially at unity or above but soon dropped to less than unity. The

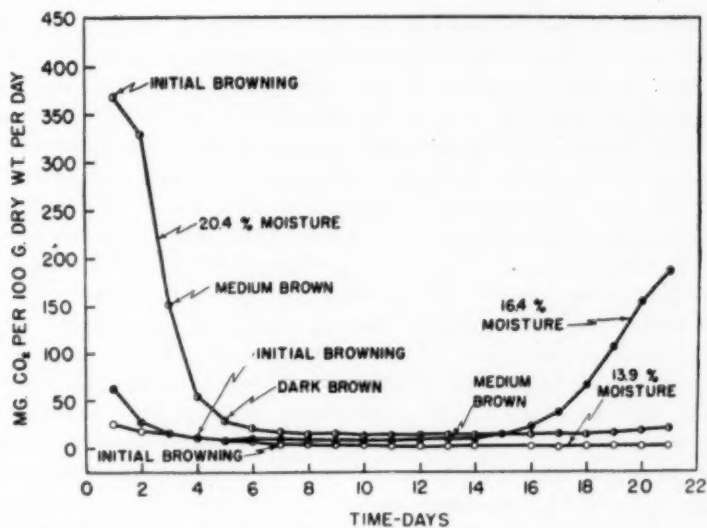


Fig. 1. Carbon dioxide production from granular wheat germ stored at various moisture contents at 40°C.

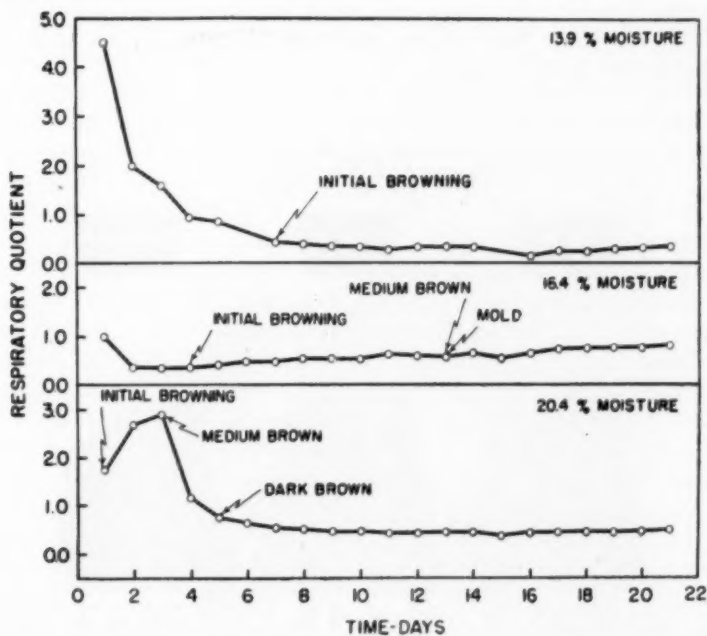


Fig. 2. Respiratory quotient values of gas exchange of granular wheat germ stored at various moisture levels at 40°C.

appearance of these samples at the end of 21 days is shown in Fig. 3.

Figure 1 records that browning was apparent in the germ sample containing 20.4% moisture 1 day after initiation of the respiration experiment, and that by only the fifth day the color had intensified to dark brown. Evidence of fungal growth in this sample did not appear, however, until a slightly elevated respiratory rate commenced on the 19th day. This was confirmed at the termination of the experiment by the appearance of one small fleck of white mold in the upper part of the sample (Fig. 3). The fact that more mold growth developed at 16.4% moisture content than at 20.4%, as indicated by the trends in the respiration curves, and in the appearance of the samples, might be considered unusual. In this case, products of the preliminary browning may have been inhibitory to fungal growth. The onset of mold growth on the sample with 16.4% moisture was evidenced on the thirteenth day by an increase in carbon dioxide evolution. Browning, however, was easily detectable on the fourth day. The sample with only 13.9% moisture showed no evidence of mold growth throughout the experiment, yet browning was evident on the seventh day. On both samples of 16.4% moisture content visible mycelia were all concentrated on the side of the flask facing the light. Gray, white, and green molds were growing on both of these samples when the experiment was terminated.

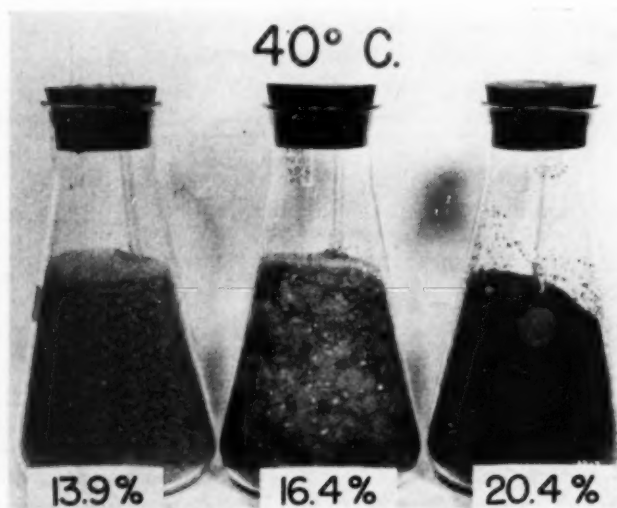


Fig. 3. Appearance of wheat germ samples stored at various moisture levels for 21 days at 40°C.



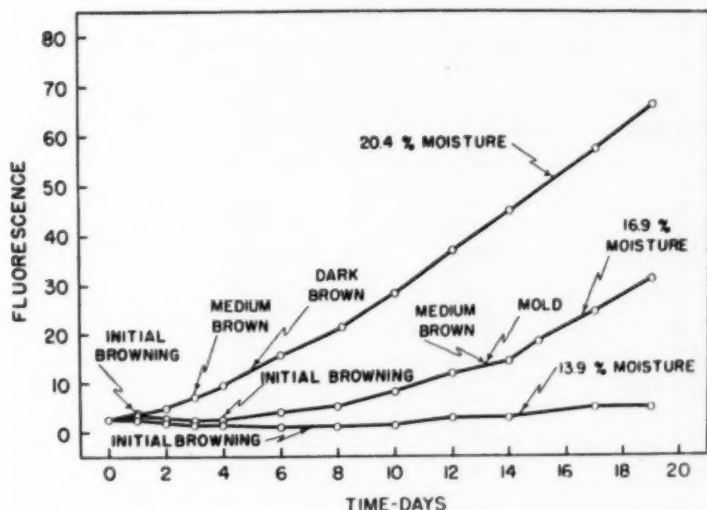


Fig. 4. Change in fluorescence of extracts of granular wheat germ stored at various moisture contents at 40°C.

The fluorescence of extracts of these samples (Fig. 4) increased very little until the wheat germ had lost its yellow color and begun to brown. As the germ turned to a darker brown the fluorescence also increased. The samples of highest moisture levels darkened most rapidly and gave the highest fluorescence values. At the end of the experiment fluorescence determinations were made on very moldy and apparently nonmoldy portions of wheat germ obtained from the flasks containing the germ of 16.4% moisture. The moldy samples were from 5 to 10 fluorescence units higher than the corresponding nonmoldy germ. This indicated that mold growth may have slightly accelerated the browning of the wheat germ. The absorbancies of extracts of these samples also increased, but irregularly (Table II). The peptizable protein (Table III) decreased in a manner analogous to the increase in fluorescence and absorbance, indicating a progressive loss in protein solubility. The pH of all three samples (Table IV) decreased gradually but regularly in a somewhat similar manner, for a total decrease of almost one pH unit. The trends in the fluorescence, absorbance, peptizable protein, and pH values of the sample of 16.4% moisture were not greatly influenced by the onset of mold growth during the experiment.

At 30°C. the initial carbon dioxide evolution from the sample containing 20% moisture was only about one-third that of the similar

TABLE II  
CHANGE IN ABSORBANCE OF EXTRACTS OF GRANULAR WHEAT  
GERM STORED AT VARIOUS MOISTURE CONTENTS AT 40°C.

Storage Time	Absorbance (245 mμ) — % Moisture		
	13.9	16.4	20.4
<i>Days</i>			
0	0.74	0.74	0.74
1	.78	0.78	0.82
2	.78	0.79	0.89
3	.78	0.78	0.94
4	.68	0.76	0.90
6	.70	0.78	0.97
8	.81	0.91	1.18
10	.83	0.94	1.24
12	.73	0.88	1.19
14	.72	0.88	1.26
15	...	0.95	...
17	.90	1.07	1.51
19	.90	1.09	1.57
21	0.76	0.91	1.42

TABLE III  
CHANGE IN PEPTIZABLE PROTEIN OF GRANULAR WHEAT GERM  
STORED AT VARIOUS MOISTURE CONTENTS AT 40°C., AS  
INDICATED BY ABSORBANCE OF  $K_2SO_4$  EXTRACTS

Storage Time	Absorbance of $K_2SO_4$ Extracts (530 mμ) — % Moisture		
	13.9	16.4	20.4
<i>Days</i>			
0	0.58	0.58	0.58
1	...	...	.58
2	.61	.58	.49
3	...	.56	.46
4	.60	.56	.41
6	.60	.51	.36
8	.60	.49	.32
10	.58	.46	.30
12	.57	.42	.26
14	.53	.38	.23
15	...	.38	...
17	.52	.32	.20
19	0.51	0.28	0.20

sample at 40°C. Without exception visible browning preceded increases in respiratory rate indicative of initiation of mold growth. At the lower temperature, other indices of browning such as increases in fluorescence and absorbance, and decreases in peptizable protein and pH, developed later and to a considerably lesser extent.

At 52°C. the initial carbon dioxide production from the high moisture samples was greater than that at 40°C., but dropped very

TABLE IV  
TREND OF pH OF EXTRACTS OF GRANULAR WHEAT GERM  
STORED AT VARIOUS MOISTURE LEVELS AT 40°C.

Storage Time	pH - % Moisture		
	13.9	16.4	20.4
<i>Days</i>			
0	6.44	6.46	6.47
1	6.43	6.40	6.38
2	6.38	6.30	6.33
3	6.28	6.18	6.27
4	6.24	6.13	6.27
6	6.15	6.04	6.22
8	6.07	6.00	6.16
10	5.97	5.97	6.10
12	5.81	5.93	6.01
14	5.78	5.92	5.97
16	...	5.93	...
17	5.73	5.88	5.84
19	5.67	5.75	5.78
21	5.60	5.68	5.68

rapidly. The respiration data indicated no evidence of mold activity, and the browning, which was more intense than that at the lower temperatures, was directly proportional to the moisture content.

*Inhibition of Browning by Sodium Bisulfite During Storage of Wheat Germ in Respirometers at 40°C.* According to Hodge and Rist (7), a characteristic of nonenzymatic browning is its inhibition by sodium bisulfite. A storage trial at 40°C. was therefore undertaken to determine the effect of this reagent on the characteristic browning of moist wheat germ in relation to other criteria of deterioration including respiration, mold growth (as indicated by respiratory characteristics), oxygen uptake, respiratory quotient, fluorescence, absorbance, and loss in protein solubility as indicated by absorbance of potassium sulfate extracts.

Wheat germ samples were conditioned to approximately 19% moisture content with distilled water and sodium bisulfite solution in such a manner that the resulting samples contained 0, 1, and 5% sodium bisulfite on the basis of the dry weight of germ. On analysis, the sample with 5% bisulfite contained 19.6% moisture while the other two samples contained 18.5% moisture. Duplicate sample pairs of the three treatment levels were stored in the respirometers at 40°C. At intervals in the course of the experiment, aliquots from one sample of each bisulfite level were removed, while gas exchange data were obtained daily on the remaining three undisturbed samples.

Data for the carbon dioxide output appear in Fig. 5.

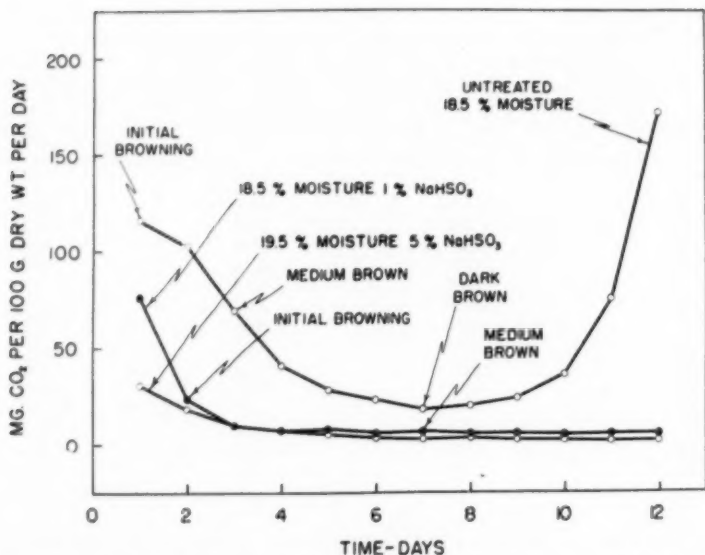


Fig. 5. Carbon dioxide production from moist granular wheat germ treated with various quantities of sodium bisulfite, stored at 40°C.

The untreated moist germ exhibited the typical high initial rate of carbon dioxide release followed by a leveling off in respiration, probably indicative of the basic respiration of the germ material. Following the sixth day, mold growth commenced as indicated by the respiration curve and developed at an increasing rate, but severe browning appeared before the evidence of mold growth in this sample. The appearance of the damp control and bisulfite-treated samples at the end of the trial in comparison with dry, nontreated germ is shown in Fig. 6.

At the 1% level of bisulfite treatment a marked suppression of initial carbon dioxide release occurred in contrast to the control. By the third day the respiration had settled down to a constant rate somewhat below that of the untreated germ. Obviously no mold growth occurred. Browning did set in, although considerably later and to a lesser degree than in the untreated sample. At the 5% level of bisulfite treatment, the initial release of carbon dioxide was further suppressed. The equilibrium value for this respiration is apparently that of the germ material uncomplicated by mold respiration. This leveling off appeared on the third day and the rate remained constant throughout the trial, at a value only slightly less than that of the sample with 1% bisulfite treatment. Thus on the sixth day the respira-

tory rate of the untreated germ was 18 mg. carbon dioxide per 100 g. of dry weight, that of the 1% bisulfite treated sample was 6.7 mg., and the 5% treatment yielded 2.6 mg. Browning, however, appeared to be virtually completely inhibited by the 5% bisulfite treatment.

The suppression of the normal high initial rate of carbon dioxide evolution from wetted germ by bisulfite was accompanied by entirely different respiratory quotient trends from those previously noted for

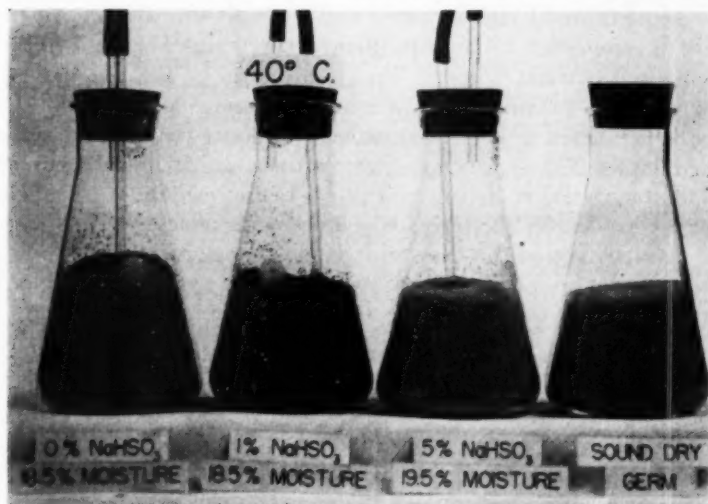


Fig. 6. Appearance of moist wheat germ samples treated with various quantities of sodium bisulfite and stored for 12 days at 40°C., in comparison with a dry untreated sample.

TABLE V  
RESPIRATORY QUOTIENT VALUES OF GAS EXCHANGE OF MOIST  
GRANULAR WHEAT GERM (APPROXIMATELY 19% MOISTURE)  
TREATED WITH SODIUM BISULFITE AND STORED AT 40°C.

Storage Time	Respiratory Quotient		
	Control	1% NaHSO <sub>3</sub>	5% NaHSO <sub>3</sub>
Days			
1	0.56	0.37	0.09
2	0.97	.12	.11
3	1.31	.15	.14
4	1.13	.29	.21
5	0.96	.39	.29
6	0.77	.36	.32
7	0.70	.43	.32
8	0.70	.40	.45
9	0.69	.34	.27
10	0.72	.31	.20
11	0.79	.32	.20
12	0.87	0.53	0.40

freshly wetted germ. The values for R.Q. in Table V indicate the usual elevation which occurs with untreated germ in the early stages, and the falling off later to normal values indicative of germ and mold respiration. Addition of bisulfite, however, causes a very marked lowering of the normal R.Q. in this early phase. Thus on the third day when the R.Q. of the untreated germ was 1.3, that of the 1% treatment level was 0.15 and that of the 5% level was 0.14. This suggests that the high initial rate of carbon dioxide evolution which ordinarily is not accompanied by an equivalent oxygen uptake (high R.Q.) is enzymatic in nature.

The visual browning of the untreated and 1% bisulfite-treated germ is paralleled by a marked increase in fluorescence of extracts of these samples (Fig. 7) which did not occur in the 5% bisulfite-treated sample. Similarly, as shown in Table VI, the bisulfite treatment inhibited considerably the increase in absorbancy of acid extracts which is a characteristic of the browning phenomenon. At the 1% level of treatment, however, the fluorescence was not reduced to the same relative extent as was absorbance.

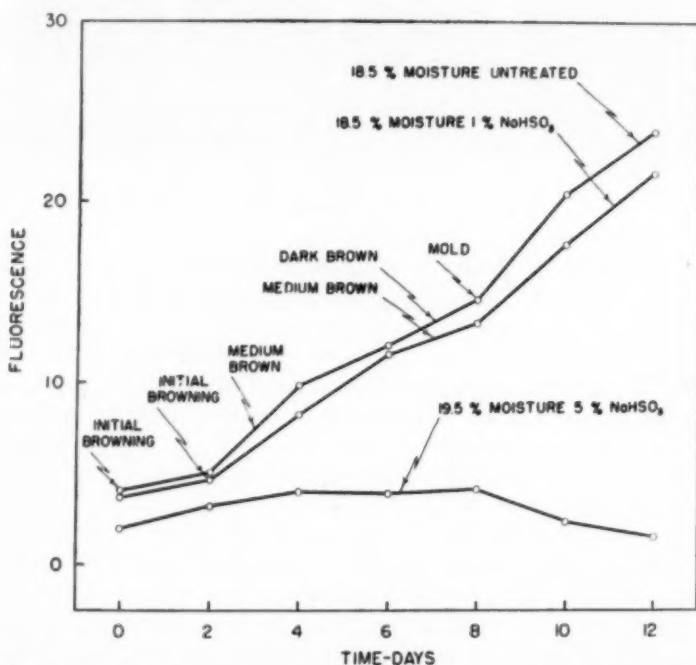


Fig. 7. Change in fluorescence of extracts of moist granular wheat germ treated with various quantities of sodium bisulfite, stored at 40°C.



TABLE VI  
CHANGE IN ABSORBANCE (245 m $\mu$ ) OF EXTRACTS OF MOIST GRANULAR  
WHEAT GERM (APPROXIMATELY 19% MOISTURE) TREATED  
WITH SODIUM BISULFITE AND STORED AT 40°C.

Storage Time  <i>Days</i>	Absorbance (245 m $\mu$ )		
	Control	1% NaHSO <sub>3</sub>	5% NaHSO <sub>3</sub>
0	0.76	0.70	0.69
2	0.82	.71	.65
4	0.89	.74	.67
6	0.96	.77	.68
8	1.02	.84	.71
10	1.06	.87	.71
12	1.04	0.81	0.65

In sharp contrast to the inhibition by bisulfite of normal increases in fluorescence and absorbance which are characteristic of browning, the usual decrease in peptizable nitrogen which accompanies browning was not inhibited by bisulfite. As shown in Table VII the drop in absorbancy at 530 m $\mu$  indicating a loss in protein solubility proceeded in all samples regardless of treatment. The sample which showed no browning (5% sodium bisulfite) exhibited the greatest loss in protein solubility. These data suggest that the increase in browning, fluorescence, and absorbancy (ultraviolet) may be inhibited by bisulfite, but that the preliminary reaction involving proteins and sugars is not affected by this reagent.

Analysis by Kjeldahl procedure of both the treated and untreated samples after storage for nitrogenous compounds soluble in 1.0 N trichloroacetic acid showed that only insignificant changes in these ma-

TABLE VII  
CHANGE IN PEPTIZABLE PROTEIN IN MOIST GRANULAR WHEAT GERM  
(APPROXIMATELY 19% MOISTURE) TREATED WITH SODIUM BISULFITE  
AND STORED AT 40°C., AS INDICATED BY ABSORBANCE  
OF K<sub>2</sub>SO<sub>4</sub> EXTRACTS

Storage Time  <i>Days</i>	Absorbance of K <sub>2</sub> SO <sub>4</sub> Extracts (530 m $\mu$ )		
	Control	1% NaHSO <sub>3</sub>	5% NaHSO <sub>3</sub>
0	0.60	0.61	0.59
2	.57	.52	.45
4	.51	.47	.40
6	.47	.45	.38
8	.42	.41	.35
10	.39	.39	.34
12	0.39	0.38	0.34

terials had occurred. Thus it appears that liberation of amino acids, short chain peptides, or other protein degradation products soluble in trichloroacetic acid is not involved in the preliminary phases of germ browning and consequently that the normal decrease in peptizable protein is not due to protein hydrolysis.

### Discussion

Results of the present study strongly favor the conclusion of Cole and Milner (4) that the brown pigment characteristic of sick wheat which forms in wheat germ is a product of a nonenzymatic browning reaction of the Maillard type between reducing sugars and nitrogenous compounds. This is indicated by the inhibition of normal browning by water-extraction of germ, the characteristic increase in fluorescence and absorbancy of acid extracts, and decreases in pH and peptizable protein.

Mold growth, as detected by respiratory changes in fresh germ stored at constant temperature and moisture content, invariably appeared after browning had already begun, even at temperatures and moisture values and within storage periods favorable to mold growth. It is concluded that the brown color of the germ which is the primary criterion of sick wheat is not caused by fungi but arises from non-biological reactions. Increases in moisture content or temperature either individually or simultaneously favor the appearance of the browning reaction. It appears that the source of these necessary environmental factors is immaterial. Under natural conditions it is probable, however, that elevation of temperature and moisture content in grain storage bins due to the respiratory activity of storage fungi indigenous to the grain is a factor in the promotion of germ deterioration.

The respiration measurements also clarified certain characteristics of the browning reaction in germ. Thus, in the storage experiment at 30°C., the sample with 13.9% moisture which exhibited no detectable browning, no mold growth, and no significant increase in absorbancy and fluorescence, tended toward a respiratory quotient of 1.0 as the storage progressed. On the other hand, the R.Q. of the sample with 15.6% moisture continued to fall steadily to values as low as 0.4 which were attained on the ninth day when browning became evident. A subsequent upturn in respiration signaled the advent of mold growth. At 20% moisture content, the early appearance of mold growth prevented the drop in R.Q. values which appears to be associated with browning. The data for the experiment conducted at 40°C. (Fig. 2) also clearly demonstrate the typical low R.Q. values associated with

browning (0.5 or less) and the tendency of mold growth, which appears after the onset of browning, to raise the R.Q. toward unity. These low R.Q. values, characteristic of the browning reaction, indicate that carbon dioxide evolution is proceeding less rapidly than oxygen uptake by a factor of about one-half. Similar low R.Q. values were noted by Milner and Geddes (12) in the spontaneous adiabatic heating of soybeans during the chemical stage of heating at temperatures just beyond the thermal death point of molds whose respiration causes the preliminary heat production. Some information exists concerning the probable mechanism of carbon dioxide evolution in browning systems (6), but considerable clarification of this mechanism in relation to oxygen uptake and heat production is needed. That a nonbiological browning reaction appears to be the strongly thermogenic agency responsible for advanced spontaneous heating of most stored agricultural materials has recently been proposed (13).

Of interest in this study was the indication of marked change in protein solubility in germ accompanying the onset of browning. Values for peptizable nitrogen always decreased apparently in direct relation to the increase in fluorescence and absorbancy (ultraviolet) of extracts. Hydrolysis of protein does not appear to be involved. This proves the major dependence of the browning reaction in wheat germ on the protein constituents. The present study demonstrated clearly, however, that the chromogens and fluorogens which are developed in, and are characteristic of, this browning were formed at a stage *following* the initial reaction with proteins. This was apparent when sodium bisulfite inhibited the development of the typical fluorescence rather strongly, the absorbancy of extracts considerably, but the characteristic decrease in peptizable protein not at all.

The nonenzymatic browning mechanism proposed by Hodge and Rist (7), involving the formation of a N-substituted 1-amino-1-deoxy-2-ketose through an Amadori rearrangement of the N-substituted glycosylamine formed by the interaction of amino compounds with reducing sugars, appears to explain these reactions adequately. The N-substituted 1-amino-1-deoxy-2-ketose would be complexed readily by bisulfite as are most aldehyde and ketose compounds. Thus color development is blocked. Such a mechanism having been invoked, it may be seen why, in the present study, nonpeptizable protein would decrease as long as conditions prevailed favoring the initial sugar-protein reaction, but dark brown compounds with characteristic absorption and fluorescence properties would not be produced.

Other investigations (6) suggest that fluorogens are precursors of the brown pigments but are not identical with them. In the presence

of bisulfite, color formation could be inhibited yet fluorogens may appear.

The phenomenon of an initially strongly elevated but decreasing rate of evolution of carbon dioxide observed in freshly wetted germ parallels similar observations with soybeans (11) and peas (5). The normally high R.Q. accompanying this initial gas exchange suggests that it originates from highly oxidized substrates such as carboxylic acids which have accumulated in the dormant embryo. The suppression of this carbon dioxide evolution by sodium bisulfite indicates inhibition of an enzymatic mechanism. Baldwin (2) proposes that bisulfite complexes certain aldehydic respiratory intermediates which are normally utilized for carbon dioxide production. This hypothesis, which serves to explain the suppression of carbon dioxide production, when combined with the observation in the present study that oxygen uptake is apparently not similarly affected, rationalizes the sharply reduced R.Q. values characteristic of bisulfite-treated germ.

#### Acknowledgments

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## IMPROVEMENT OF FLOUR MIXING CHARACTERISTICS BY A STEARYL LACTYLIC ACID SALT<sup>1</sup>

J. B. THOMPSON AND B. D. BUDDMEYER<sup>2</sup>

### ABSTRACT

Farinograph and baking tests showed that the inclusion of calcium stearyl-2 lactylate at levels of about 0.5% (flour basis) in doughs increased mixing tolerances without materially altering the optimum mixing times.

Repeated attempts, in some cases with a measure of success, have been made by cereal technologists to modify the mixing properties of wheat flour through the alteration of the characteristics of the gluten proteins. Some studies of this nature have been made to gain an understanding of the underlying causes of the observed differences in baking quality between otherwise comparable flours. Others have been conducted because of the obvious practical potentialities for improvement of bakeshop practice if the mixing characteristics of flours could be altered and controlled.

Various methods have been employed to modify the mixing characteristics of flour, including heat treatment (2), solvent extraction to remove some of the lipid materials (1, 3), and the addition of oxidizing improvers. The only procedures which have been widely adopted for modifying the mixing characteristics have involved the addition of small quantities of active agents to flour, which accomplish the result through chemical alteration of the flour proteins, the destruction of deleterious reducing materials, or both. The use of these oxidizing and maturing agents and their effects have been studied by many workers in the field.

Practically, the modification of the mixing characteristics of flour by additions of traces of suitable reagents has been the most attractive and the search for such agents has continued.

Swanson and Andrews (5, 6) determined the effects of a number of anionic surface-active agents, particularly a commercial sodium dioctyl sulfosuccinate, upon the mixing characteristics of flours in the mixograph. Many of the agents markedly increased the mixing time of flours, but attempts to correlate the activity of the materials in dough with their surface tension-depressing properties were futile. From the behavior of flours having different protein contents, when mixed with traces of the surface-active agents, the authors concluded

<sup>1</sup> Manuscript received December 30, 1953.

<sup>2</sup> C. J. Patterson Company, Kansas City, Missouri.



that there was interaction between the agent and protein causing some type of selective denaturation of the latter.

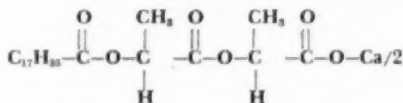
Swanson and Johnson (7) employed two surface-active agents, sodium dioctyl sulfosuccinate and sodium lauryl sulfate, in baking studies at levels of less than 0.25%. The sodium lauryl sulfate was entirely unsatisfactory, while the sodium dioctyl sulfosuccinate increased the mixing time required to obtain bread comparable in quality with the controls. The latter material, however, did increase the volume of test loaves.

Sullivan (4) studied the effects of a variety of cationic, anionic, and nonionic surface-active materials upon the mixing characteristics of flour in the farinograph. Of these materials, only those of the anionic type were effective in increasing the mixing time and tolerance. Nacconal, a commercial sodium alkyl acyl sulfonate, used in the farinograph doughs at levels of 0.1% to 1.0%, was particularly effective. It is noteworthy that Sullivan found that the sodium soaps of higher fatty acids produced qualitatively similar effects. She concluded that these apparent improvements in mixing time and tolerance were the result of reaction between the detergents and flour proteins. However, baking experiments did not reveal these same improvements in mixing tolerance or dough-handling characteristics.

While some understanding of the causes of variations in tolerances and mixing times of flours has been gained, these studies have offered no method for modifying these characteristics. The purpose of the present paper is to describe experiments using a stearyl lactic acid derivative which appeared to have merit for such a use.

### Materials

The compound employed in this investigation was a mixture of the calcium salts of a homologous series of stearyl lactic acids<sup>a</sup>, the following being its average structure:



Physically, the calcium stearyl-2 lactylate used was a fine powder not unlike flour in appearance. In this form it was free flowing, non-hygroscopic, and quite stable. This powder was employed in both farinograph and baking tests.

The calcium stearyl-2 lactylate powder was only sparingly soluble

<sup>a</sup> VERV-Ca, product of C. J. Patterson Company, Kansas City, Mo.

in water, but water seemed to be adsorbed upon it. Upon stirring, it dispersed rather thoroughly, but upon heating of the mixture, the compound separated as a plastic mass on the surface of the water. The pH of a 2% dispersion of the powder in water was 4.7; such a pH would cause no adverse hydrogen ion effects in a dough.

The salt, upon ashing by usual procedures with nitric acid as an aid, yielded an ash of CaO. Titration of this ash indicated that the salt contained 4.38% calcium. The theoretical amount based on the average structure given is 4.48%.

The flour employed was a commercial baker's patent milled from hard red winter wheat; it had 11.2% protein and 0.44% ash on a 14% moisture basis.

### Methods

*Mixing Curves.* The Brabender Farinograph with large bowl was used in the preparation of the mixing curves. The farinograms were prepared using 300 g. of flour on a 14% moisture basis. When calcium stearyl-2 lactylate was employed, it was simply added at a predetermined level based on the flour and blended in by running the machine for several minutes without water.

After the normal absorption required to give a peak viscosity of 500 B.U. was determined by titration, this quantity of water was kept constant for the remainder of the mixing tests regardless of the amount or effects of the calcium stearyl-2 lactylate added. Two criteria of mixing performance were obtained by measurements of the curves: 1) mixing time—the time in minutes required to obtain peak consistency readings; and 2) a mixing tolerance index—the decrease in consistency expressed in Brabender units which occurred upon mixing 5 minutes beyond the time required to obtain maximum consistency.

*Bake Tests.* Breads were made for this study, using the sponge and dough procedure and the following lean but typical commercial formula:

	<u>Weight</u>
	<u>g.</u>
Flour	700.0
Water	430.5
Yeast	14.0
Yeast food-Fermaloid	3.5
Nonfat milk solids	21.0
Sugar	35.0
Lard	14.0
Salt	14.0
VERV-ca	3.5

The sponges, containing 420 g. of flour, 239 g. of water, with the

yeast and yeast food, were mixed 1 minute at low speed followed by 1 minute in second speed in a jacketed McDuffee bowl on a Hobart A-200 Mixer. The sponges were fermented 4.5 hours at 84°F. (28.89°C.) and 85% relative humidity.

To prepare the doughs, the remainder of the dry ingredients, including the calcium stearyl-2 lactylate at 0.5%, if used, and the water were placed in the mixer. The ingredients were mixed 3 minutes at low speed while the fermented sponge was being added in four parts.

The speed was increased to second and maintained for the desired periods, which were 1, 3, 5, 7, 10, and 13 minutes. The temperature was controlled through the ice-water jacket and the doughs came out with a temperature of 80°F. (26.67°C.).

The doughs were given a floor time of 35 minutes and divided to form two 480-g. dough pieces. These were made up in a normal manner, proofed to equal volume, and baked 25 minutes at 450°F. (232.2°C.).

The bread was permitted to cool for 1.5 hours, and volumes of the loaves were determined by the rape seed displacement procedure. After 24 hours of storage in polyethylene bags, the bread was cut and scored. The quality scores given are relative and subjective.

### Results and Discussion

The farinograms in which calcium stearyl-2 lactylate was used at 0%, 0.25%, 0.50%, and 1.00% based on the flour, are reproduced in Fig. 1. The significant data read from them are summarized in the following table:

Calcium stearyl-2 lactylate, flour basis	Peak time	Mixing tolerance index, Brabender Units
%	min.	
0.00	6.5	40
0.25	6.5	30
0.50	5.0	20
1.00	10.5	15

The calcium stearyl-2 lactylate had an anomalous effect on the peak time as read from these farinograms; there was no evident effect at 0.25%, whereas apparent mixing time was decreased at 0.50% and increased at 1.00%. The significance of these variations is not definitely known, but possibly they indicate insensitivity in the method rather than any such extreme effects of the compound on actual mixing time.

These curves indicated that the calcium stearyl-2 lactylate at the levels used had a pronounced ability to increase mixing tolerance as

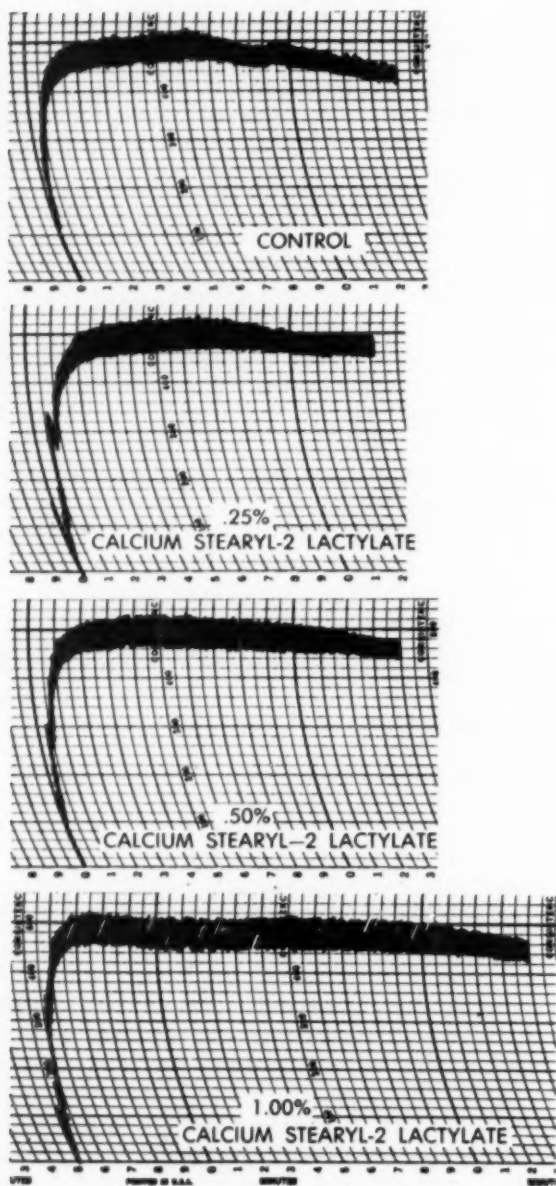


Fig. 1. Effect of calcium stearyl-2 lactylate on farinogram mixing characteristics.

shown by the decreased mixing tolerance index. This effect was generally proportional to the amount of the calcium salt employed, a maximum effect being approached at 0.50% based on flour weight.

The mixing time series, in general, verified those predictions for functionality made by the farinograph tests. The data obtained from these baking tests are summarized in Table I.

TABLE I  
MIXING TIME SERIES: EFFECT OF 0.5% CALCIUM STEARYL-2 LACTYLATE  
ON LOAF VOLUME AND BREAD QUALITY

Mixing Time	Loaf Volume		Quality Score*	
	Calcium Stearyl-2 Lactylate	Control	Calcium Stearyl-2 Lactylate	Control
Min.	cc.	cc.		
13	2925	2825	78	75
10	2975	2950	79	76
7	3200	3175	84	80
5	3300	3150	85	81
3	3400	3300	86	85
1	3275	3300	86	85

\* Relative score using the system of the American Institute of Baking.

The calcium stearyl-2 lactylate at 0.50% based on the flour increased the mixing tolerance to both under- and overmixing. Without the additive, the optimum mixing time was from 1 to 3 minutes, with definite overmixing evident at 5 minutes. With the calcium salt, quality bread was obtained within the mixing time range of 1 to 7 minutes; the results would indicate that any change in optimum mixing time was slight.

The bread containing the calcium stearyl-2 lactylate was definitely of higher quality than that without it, at each mixing time, and the trend was toward a slightly greater volume than the comparable control. The material had the unpredicted effect of producing bread with a finer grain and a distinctly more tender crumb.

The stearyl lactic acid product, like the anionic surface-active agents employed by Sullivan (4) and others, altered the shape of the flour farinograms to indicate increased tolerance and mixing time. Unlike the surface-active agents, baking tests substantiated these results. The lack of correlation between surface activity and mixing time effects reported by Swanson and Andrews (5) was observable in these tests. The low solubility of the calcium salt used would seem to preclude the possibility of surface activity being more than a minor fac-

tor in accounting for its effects.

The calcium ion alone is not responsible for the magnitude of the action of calcium stearyl-2 lactylate; the anionic portion of the molecule is the active part. The alteration of flour quality is probably due to a colloidal binding of this anion to the flour protein.

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# THE ALBUMIN AND GLOBULIN CONTENTS OF WHEAT FLOUR AND THEIR RELATIONSHIP TO PROTEIN QUALITY<sup>1</sup>

JAMES W. PENCE, N. E. WEINSTEIN, AND D. K. MECHAM

## ABSTRACT

The soluble-protein contents of 32 flours of widely varying type and baking quality were found to range from about 13 to about 22% of the total flour protein, with a slightly greater proportion of albumin than globulin present, on the average. Both the albumin and globulin contents, as well as the ratio of albumins to globulins, varied significantly among the flours. The amounts of soluble proteins were found to increase directly with total flour protein, but the relationship became inverse when they were expressed as percentages of total protein.

The coefficient of regression of loaf volume on flour protein for each of the flours was used as an estimate of the baking quality of the protein systems of the various flours, although some difficulty was encountered in determining statistically reliable coefficients for several of the flours. The different levels of protein required were obtained by fortification of each flour with its own gluten and water solubles and by dilution with its own starch.

Neither the amounts of the soluble proteins nor the proportions of soluble to gluten protein are correlated significantly with protein quality as measured by the regression lines. The ratio of albumins to globulins contained by the flours, however, is correlated with protein quality ( $r = +0.60$ ) beyond the 1% point of significance. Although the total protein content of the flour is also correlated very significantly with protein quality, the partial correlation of albumin-to-globulin ratio with protein quality (obtained by holding flour protein constant) retains a high degree of significance.

The observations of Finney (4) and of Pence, Elder, and Mecham (10) have indicated that the soluble constituents of flour might be more closely associated with variations in baking characteristics among flours than had been previously realized, but knowledge concerning these constituents is quite limited. The protein contained by the mixture of soluble constituents was found by Pence, Elder, and Mecham (10) to be responsible for the greatest part of the effect exerted by this fraction on the loaf volume of reconstituted doughs prepared from several flours under the experimental conditions employed. Continued investigation has shown this protein fraction to include both albumins and globulins (9). The albumins, particularly, were found to be a complex group of closely related components (8, 9). However, a method of analysis, based on certain chemical characteristics, was devised for determination of the total amounts of albumin and globulin proteins contained by various flours (11). The present paper describes the ap-

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plication of this method to a series of 32 flours which differ widely in type and baking characteristics. The analytical results are then compared with baking data, also obtained on the flours, in an effort to discover a relationship between albumin and globulin composition and baking performance.

### Materials and Methods

The flours used were unbleached, straight-grade flours experimentally milled from pure-variety samples of wheat. Seventeen of the wheats were obtained during the 1946 crop year from locations within major producing areas of the entire country, and represent major commercial types and a wide range of baking characteristics; 13 of these flours are described in detail elsewhere (2). Ten flours were from five varieties of wheat grown in two locations (Moro and Pendleton, Oregon) in the Pacific Northwest during the 1948 crop year, and the remaining five flours were from the same varieties grown at one location (Moro, Oregon) during the 1949 crop year. Baking data were obtained on the flours when received, and the remainders were immediately placed in storage at  $-10^{\circ}\text{F}$ . in tightly closed metal cans. Subsequent tests have shown that the baking performance of the flours has not deteriorated in the intervening period.

Baking experiments were conducted by methods described previously (10), using A.A.C.C. standard pup-loaf, high-form pans. Albumin, globulin, and nonprotein nitrogen contents of the flours were determined by the methods described in a preceding paper (11). Gluten nitrogen contents were calculated as the difference between the total nitrogen content and the sum of the albumin, globulin, and nonprotein nitrogen contents.

### Albumin and Globulin Determinations

The albumin and globulin contents found for the 32 flours are given in Table I along with the values obtained for nonprotein nitrogen and those calculated for gluten. Determinations were run in duplicate using 0.5M sodium chloride extracts of the flours. With 17 of the flours, however, duplicate determinations of globulin contents had been obtained separately, using 0.5M potassium sulfate extracts, and these values are included in the mean values shown.

The differences between duplicate values were rather large for some of the flours, reaching a maximum of  $\pm 12\%$  of the mean in those instances for which means of duplicate values are reported in Table I. Differences up to  $\pm 15\%$  were obtained between duplicate

TABLE I  
THE PROTEIN COMPOSITION AND QUALITY EVALUATION OF FLOURS OF WIDELY  
VARYING TYPE AND BAKING CHARACTERISTICS

Variety <sup>a</sup>	Wheat Type	Flour Protein (N×5.7) <sup>b</sup>	NPN <sup>c</sup>	Albumin <sup>d</sup>	Globulin <sup>d</sup>	Gluten <sup>d</sup>	Albumin-Globulin Ratio	Regression Coefficient* (Baking Quality)	cc/100
1. Hymar	Club	5.7	2.5	9.7	10.4	77.4	0.93	30.5	18.3
2. Elgin, 48P	Club	6.2	2.2	9.4	10.5	77.9	0.91	38.9	134.3†
3. Rio, 48P	HRW	6.9	2.0	10.6	8.7	78.7	1.24	43.0	12.4
4. Rex, 48P	White	6.9	2.2	10.9	10.5	76.4	1.04	37.4	4.3
5. Elgin, 49M	Club	7.0	3.2	8.8	9.6	77.4	0.92	38.8	42.3†
6. Elgin, 48M	Club	7.2	2.3	9.8	9.5	78.4	1.03	41.5	16.1†
7. Yorkwin	White	7.6	1.8	9.3	9.2	79.7	1.01	35.4	7.2
8. Rio, 48M	HRW	7.7	2.2	10.8	8.6	78.4	1.27	42.9	9.6
9. Rex	White	7.8	2.4	10.1	8.8	78.7	1.15	45.7	24.4†
10. Baart, 48P	White	8.2	2.2	11.5	8.8	77.5	1.32	53.3	5.9
11. Rex, 49M	White	8.3	2.7	10.7	10.0	77.6	0.97	39.9	4.2
12. Baart, 48M	HRW	8.4	2.8	8.7	7.6	81.6	1.13	47.7	37.3†
13. Baart, 48M	White	8.6	2.5	10.9	7.8	79.0	1.45	50.7	30.1
14. Parkman	S-HRW	8.7	1.6	10.7	7.8	79.9	1.37	54.5	13.5
15. Tennesse	HRW	8.8	2.1	7.5	8.5	81.9	0.88	33.5	13.6†
16. Chiefkan, 48P	HRW	8.8	1.9	8.4	7.7	82.0	1.10	50.8	23.4†
17. Hymar	Club	9.0	1.5	7.4	8.0	83.1	0.93	30.2	6.4
18. Rio, 49M	HRW	9.0	2.4	10.4	7.8	79.4	1.37	36.2	21.0
19. Goens	SRW	9.1	1.9	7.7	8.1	82.3	0.95	31.7	11.3
20. Baart	White	9.5	1.8	9.3	7.4	81.5	1.26	48.7	9.9
21. Chiefkan, 49M	HRW	9.5	2.7	8.6	7.8	80.9	1.11	43.1	12.5†
22. Rex, 48M	White	9.5	2.9	7.8	10.0	79.3	0.78	59.9	18.3
23. Chiefkan	HRW	9.6	1.6	7.5	8.3	82.6	0.90	31.2	28.9†
24. Conanche	HRW	9.7	1.7	9.8	6.8	81.7	1.44	56.9	60.8†
25. Baart, 49M	White	9.8	1.5	10.6	10.8	76.6	0.97	37.1	18.4
26. Tennesse	HRW	10.2	1.5	10.6	6.7	84.1	1.55	47.1	5.5
27. Red Chief	White	10.3	1.5	7.6	7.2	83.2	0.99	48.9	12.0
28. Pentad	Durum	11.6	2.2	6.8	7.2	83.3	0.94	44.0	12.8
29. Premier	HRS	11.7	1.4	9.3	6.3	83.0	1.48	54.0	6.1
30. Red Chief	HRW	12.9	1.3	7.9	5.9	84.9	1.34	50.8	9.9
31. Turkey	HRW	13.4	2.0	9.1	5.7	83.2	1.60	61.6	5.1
32. Thatcher	HRS	14.2	1.8	7.8	5.8	84.6	1.34	67.2	1.6
Mean		9.1	2.1	9.1	8.3	80.5	1.14	44.1	

<sup>a</sup> Figures following variety name represent crop year and growth location (Moro, Oregon, or Pendleton, Oregon).

<sup>b</sup> At 14% moisture.

<sup>c</sup> Expressed as percentage of flour nitrogen.

<sup>d</sup> Expressed as percentage of flour protein.

\* Regression of loaf volume (cc.) on flour protein (%). The figures following the coefficients denote the 95% limits of confidence. The figures marked with a dagger (†) indicate coefficients for which only three levels of protein were available for determination of the regression line.

values for the globulin contents of four flours, but these are included in the group for which four replicates were used to calculate mean values. Confidence intervals, therefore, are of the same order for all the mean values shown. The minimum significant differences (at the 5% point) between mean values for the individual flours were found to be 1.5% of the total flour protein for both albumins and globulins. An analysis of variance showed that a very highly significant variation exists among the mean values for the albumin and globulin contents of the flours.

Correlation calculations, summarized in Table II, show that the

TABLE II  
STATISTICAL COMPARISON OF VALUES FOR VARIOUS PROTEIN COMPONENTS  
WITH TOTAL FLOUR PROTEINS ( $N \times 5.7$ )

Variable Correlated With Flour Protein	Correlation Coefficient	
	Variable as % of Flour Weight	Variable as % of Flour Protein
Albumin	+ 0.71**	- 0.46**
Globulin	+ 0.45**	- 0.79**
Albumin + globulin	+ 0.83**	- 0.83**
Gluten	+ 0.99**	+ 0.75**

albumin, globulin, and gluten contents, as well as the total soluble proteins (albumin plus globulin), increase in amount with the total flour protein ( $N \times 5.7$ ). The percentage values for the soluble proteins, however, decrease with increasing total protein, illustrating that the rate of increase in actual amount of the soluble proteins is much smaller than that for the gluten proteins.

Few quantitative estimates of the albumin and globulin contents of flours are to be found in the literature, although many studies concerning the amounts of flour protein soluble in various salt solutions have been made (1). In general, relationships that were found between the soluble and total proteins of various flours in previous studies are similar to those reported in the present paper. The amounts of protein extracted from flour by solutions of such salts as sodium chloride or potassium sulfate were found in a previous study (11) to be similar to the total amounts of albumins and globulins in flour because relatively small amounts of gliadin are extracted by these particular salts. With salts such as bromides or iodides, however, significant discrepancies occur due to the large amounts of gliadin extracted along with the albumins and globulins.

### Determination of Baking Quality

Because of the definite differences found among the albumin and globulin contents of the 32 flours, it was of interest to determine whether a relationship to baking properties of the flours could be demonstrated. However, as it is commonly accepted that the protein of wheat flours is the principal factor in determining their bread-baking properties, a direct comparison of the baking performance of more than a few of the flours was not possible because of their wide variation in protein content.

Objective measurement of the baking quality of flours is difficult at best, and comparisons between flours of different protein content are especially hazardous. Finney and Barmore (5), among others, found that the relationship between flour proteins and loaf volume was linear over a wide range of protein content for flours from hard red winter and spring wheats. These workers also observed that some varieties had distinctly different lines for the regression of loaf volume on flour protein, and they concluded that variety regression lines reflected true differences in protein quality. The very highly significant correlation between the slope of such regression lines and loaf volume at a constant protein level enabled them to develop a logical method of correcting loaf volume for differences in protein content of flours from hard red winter or spring wheats.

In an attempt to overcome the obstacle of varying protein content in the present study, each flour was baked at several protein levels by the method used by Finney (4). The separate fractions of gluten, water-solubles, and starch from each flour were added back to the flour from which they were obtained to produce flours varying up to about  $\frac{20}{7}\%$  in protein content above and below the original level. The coefficient of regression of loaf volume on flour protein thus obtained was considered to be a measure of baking quality of the *protein* systems of each flour. The values obtained are included in Table I. It was felt that the slope (coefficient of regression) of the regression line would be a more reliable measure of the quality of flour protein than a corrected loaf volume, because it was desired to compare flours from different classes of wheat and because other flour constituents, such as starch, might affect loaf volume at a given level of protein when comparisons were made among the widely different types of flours.

It is to be expected that values obtained for the flours used in the present study would be lower, in general, than those reported by Finney and Barmore (5) and Fifield, Weaver, and Hayes (3) because the majority of the present samples are from flours normally considered

to be unfit for commercial bread production, either by reason of variety or owing to low protein content. The relatively high quality values obtained for the Red Chief and certain of the Chiefkan flours contrasts somewhat with the low acceptability for bread production normally associated with these flours. Other workers (6, 7), however, have observed that the baking performance of Chiefkan gluten, added to a standard flour, ranks high among those for other glutes which were obtained from flours considered of better quality. The results in Table I also show that the quality of the *protein* from flours of low protein content may often be high even though the level of protein naturally present is insufficient for the production of good bread.

The low order of reliability of many of the regression coefficients, as indicated by the confidence limits shown in Table I, results primarily from the small number of points used to determine the regression lines. With 22 of the flours, four or more levels of protein were used, but with ten of the flours only three levels were available. The variability of the baking values themselves is quite similar for all of the flours, but the decrease in the number of degrees of freedom when only three levels of protein are used for calculation of the regression line affects the statistical evaluation of the results so that three points are virtually insufficient to determine the true slope of the regression lines.

### Relation of Composition to Baking Quality

Statistical comparisons between the baking-quality values and values for various compositional features of the flours are summarized in Table III. The highly significant correlation between protein quality (as measured by the regression coefficients) and the albumin-to-globulin ratio (line 1) is of immediate interest; but the implications

TABLE III  
STATISTICAL COMPARISON OF VALUES FOR AN ESTIMATE OF PROTEIN  
QUALITY WITH PROTEIN COMPOSITION OF FLOURS

Variables Correlated		Correlation Coefficient
Albumin/globulin	× Regression coefficient*	+ 0.60**
Total flour protein ( $N \times 5.7$ )	× Regression coefficient*	+ 0.60**
Albumin/globulin	× Total flour protein ( $N \times 5.7$ )	+ 0.37*
Albumin + globulin/gluten	× Total flour protein ( $N \times 5.7$ )	+ 0.82**
Albumin + globulin/gluten	× Regression coefficient*	- 0.33
Albumin/gluten	× Regression coefficient*	+ 0.02
Globulin/gluten	× Regression coefficient*	- 0.38*

\* Regression of loaf volume on flour protein.



of this relationship are greatly influenced by the correspondingly significant correlation between protein quality and total protein (line 2). However, the albumin-to-globulin ratio is correlated with total protein to a less significant degree (line 3), and the partial correlation coefficient between protein quality and the albumin-to-globulin ratio, obtained by holding total protein constant, retains a high degree of significance ( $r = +0.511$  and  $P = <0.01$ ). These relationships are interpreted as support for the hypothesis that the soluble proteins of flour are related to the variations in baking performance observed among flours (10). The ratio of total soluble to gluten protein (line 5) appears to be unrelated to quality performance, as measured by the method used, although it is very highly correlated with total protein content (line 4).

The magnitude of the correlation coefficients obtained for both total protein and albumin-to-globulin ratio against protein quality, although highly significant, is too small for reliable prediction of the protein quality of unknown flours by means of either factor alone. This is emphasized by the several widely divergent points seen in Fig. 1 in which albumin-to-globulin ratios are plotted against regression coefficients. The various flours are identified by the number of their listing in Table I. Figure 2 shows a similar comparison between protein quality and total protein. It thus appears that flours having

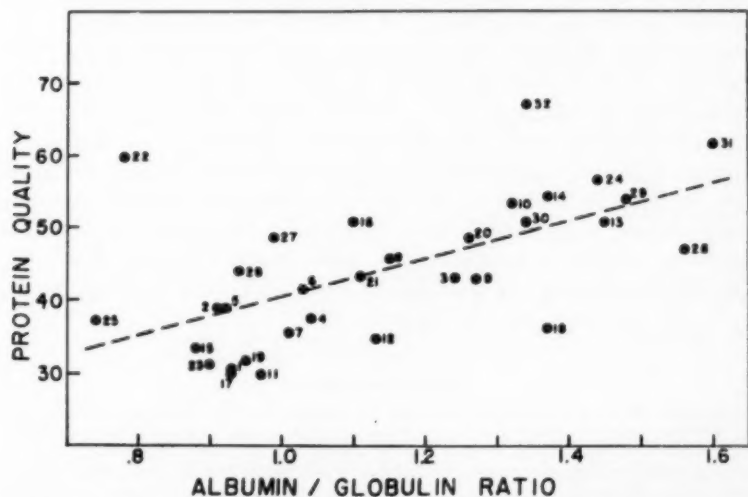


Fig. 1. The relation between albumin-to-globulin ratio and protein quality (coefficient of regression of loaf volume on protein content) for 32 flours of widely different type and baking performance ( $r = +0.60^{**}$ ).



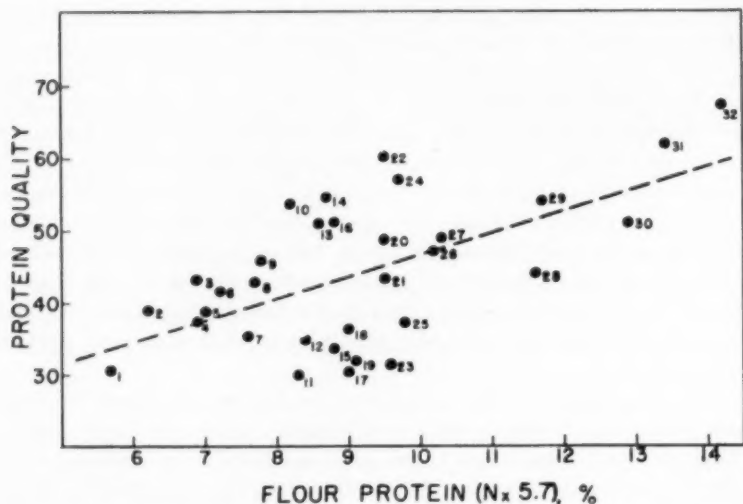


Fig. 2. The relation between flour protein ( $N \times 5.7$ ) and protein quality (coefficient of regression of loaf volume on protein content) for 32 flours of widely different type and baking performance ( $r = +0.60^{**}$ ).

high-quality protein usually have a high ratio of albumin to globulin and a naturally high level of protein.

Despite the large uncertainty regarding several individual values, the correlation illustrated in Fig. 1 is that of an unbiased sample and constitutes, at least, a strong suggestion that the ratio of albumins to globulins in flour may be related to protein quality. Certainly, the mechanisms by which soluble proteins might influence baking behavior are worthy of further investigation. Although it is difficult to imagine a logical manner in which the two groups of proteins could interact with each other so as to affect the baking performance of a bread dough, it is possible that albumin components can act in a beneficial manner in bread doughs, whereas globulins could act in a deleterious manner. The resultant of the two effects would then be reflected by the ratio of their amounts.

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## THE FLOUR FACTOR IN THE FARINOGRAPH TEST FOR EVALUATING THE BAKING PROPERTIES OF MILK SOLIDS<sup>1</sup>

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AND GASTON DALBY<sup>2</sup>

## ABSTRACT

A sample of nonfat milk solids of borderline quality for breadmaking was tested by a farinograph procedure based upon a determination of the hydration time and maximum absorption of a mixture of the milk solids with an equal quantity of wheat flour. In general, spring patents, winter patents, spring clears, and variations in chemical treatment all resulted in essentially the same final evaluation. Flour streams of very short extraction contained protein which was less resistant to the adverse effect of poor milk solids than the less highly purified streams.

The flour factor does not appear to be a serious drawback to the use of the farinograph procedure, but it must always be taken into account.

The proper evaluation of the baking properties of nonfat milk solids continues to be of commercial importance to the baker. To the cereal chemist, the action of improperly processed milk solids in a bread dough is of considerable theoretical interest, but as yet no complete

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explanation is at hand. A thorough evaluation of presently available methods for the determination of baking properties of milk solids has been given by Larson *et al.* (2).

In a commercial laboratory, the drawbacks to the experimental baking test are the time consumed in testing large numbers of samples in addition to the fact that, in a small dough, a milk tends to give generally better baking results than those of the same milk in a large commercial dough. As a control procedure, therefore, the use of the experimental baking test can permit the acceptance of milk in the bakery that is not up to satisfactory quality for commercial doughs. The farinograph procedure of Hoffman *et al.* (1) was designed to magnify the effects of milk solids on flour. This was achieved by the use of equal parts of flour and milk solids in the dough tested in the farinograph. For the flour portion used in the test dough, the proper absorption is determined on the flour alone. For the milk portion, an absorption of 65% is taken as an arbitrary level. The absorption of the mixture of flour and milk solids is therefore an average of the flour absorption and 65% used for the milk. Milk of poor quality immediately softens the dough to a marked degree while one of good quality produces a very stiff dough. By comparison with commercial results, standards for absorption and hydration period, or dough development time, have been worked out that permit specifications, based on farinograph results, to be set up. It is reasonably certain that this procedure eliminates all milks of poor baking quality, but it is admitted that it may reject some which would bake satisfactorily.

In the last several years, there has been some indication that variations in the flour were influencing the results of the test. Flour variations did not appear to affect the evaluation of either excellent or poor milks, but only that of milks of borderline quality. Such milks will give reasonably satisfactory results in the commercial shop but do not produce bread of the very best quality. Shop conditions may be adjusted to permit the successful use of a borderline milk. Total absorption may be decreased, yeast may be increased, and fermentation times and temperatures adjusted. In large-scale operations such adjustments are not desirable.

The experiments reported here were undertaken to determine the extent to which variations in flours may influence the evaluation of nonfat milk solids of borderline quality.

#### Materials and Methods

A well-mixed sample of roller process nonfat milk solids of border-

line quality was employed. Several commercially milled flours were used, including spring clears (about 15.0% protein and 0.70% ash), winter patents (about 11.5% protein and 0.43% ash), and spring wheat patent flour (11.6% protein and 0.43% ash), with and without chemical treatment with various combinations and levels of chlorine dioxide and benzoyl peroxide, and various flour streams from a spring wheat mill.

Farinograph tests were made of the milk solids with each of the flours by the procedure described by Hoffman *et al.* (1). The flour absorption (F) is first determined by the farinograph in the usual manner. One hundred and fifty grams, each, of flour and milk solids are then placed in the farinograph and an amount of water equal to  $(F + 65)/2$  is added and the mixer operated to develop the dough. The time required for the dough consistency to attain a Brabender consistency of 500 units is recorded as the hydration period. During the first few minutes the consistency may exceed 500 units owing to incomplete mixing, and small increments of water are added, if necessary, to hold the consistency at the 500 line until maximum absorption is reached. If X is the total amount of water required, the absorption of the milk solids equals  $2(X - F/2)$ .

### Results and Discussion

The data obtained through the use of the different types of flours are recorded in Table I. The milk sample gave a calculated absorption exceeding 70%, with all flours except the spring patents Nos. 1A, 1B,

TABLE I  
EVALUATION OF NONFAT MILK SOLIDS OBTAINED WITH VARIOUS FLOURS

Flour	Flour Absorption	Hydration Period	Calculated Milk Absorption
	<i>%</i>	<i>min.</i>	<i>%</i>
Spring clears, 3 samples			
Minimum	66.0	14.5	75.0
Maximum	66.2	16.5	76.0
Spring patents			
No. 1A	63.8	14.0	68.0
No. 1B	62.5	13.5	67.5
No. 1C	62.5	10.5	69.5
No. 2	64.0	9.0	72.0
No. 3	61.8	9.5	71.5
Winter patents, 7 samples			
Minimum	60.8	6.0	72.5
Maximum	62.0	11.0	77.0

and 1C, comprising one brand of flour from three mills.

The results with a spring wheat patent flour subjected to various degrees of chemical treatment in Table II suggest that chemical treatment of a flour does not affect the evaluation of a milk sample.

TABLE II  
EFFECT OF CHEMICAL TREATMENT OF A SPRING WHEAT FLOUR ON ITS UTILITY FOR  
EVALUATION OF THE NONFAT MILK SOLIDS

Flour Treatment	Flour Absorption	Hydration Period	Calculated Milk Absorption
	%	min.	%
None	64.0	8.5	72.5
0.5 oz. Benzoyl peroxide <sup>a</sup>	64.5	8.0	73.0
Low chlorine dioxide <sup>b</sup> ; no benzoyl peroxide	64.0	8.5	72.0
High chlorine dioxide; no benzoyl peroxide	63.0	8.0	73.0
High chlorine dioxide, 0.3 oz. benzoyl peroxide	64.0	8.5	72.0

<sup>a</sup> Benzoyl peroxide is given in terms of oz. per cwt. of flour.

<sup>b</sup> The mill that prepared the samples did not specify exact levels. "Low" chlorine dioxide would be in the range of 0.1 g. per cwt. of flour. "High" chlorine dioxide would be 0.6 to 0.7 g. per cwt. of flour.

The data obtained with mill stream flours from two sources given in Table III indicate that the protein from the most highly purified streams reacts with milk to give a lower absorption than that of the streams of longer extraction.

TABLE III  
EVALUATIONS OF NONFAT MILK SOLIDS OBTAINED WITH FIVE SERIES OF MILL STREAMS  
(SPRING WHEAT)

Stream	Ash	Protein	Flour Absorption <sub>s</sub>	Hydration Period	Calculated Milk Absorption
	%	%	%	min.	%
Mill No. 1					
Patent	0.37	10.8	68.5	11.5	66.5
Clear No. 1	0.66	13.3	63.5	7.0	76.5
Clear No. 2	0.65	14.1	72.5	10.0	75.0
Clear No. 3	0.92	12.4	74.0	11.0	72.0
Mill No. 2					
Stone Stock	0.36	11.1	70.8	11.0	68.5
1st Middlings	0.38	11.0	68.0	9.5	69.0
1st Clear	0.71	13.2	72.0	9.5	75.0
2nd Break	0.56	15.1	68.8	13.5	76.0
Coarse sizings	0.61	12.5	69.0	11.0	70.0

The second break flour, which contained the highest protein content of all the streams, gave the highest absorption and a further series of tests was made in which this flour was diluted with corn starch to two lower protein levels. The results are recorded in Table IV.

TABLE IV  
EFFECT OF DILUTION OF SECOND BREAK FLOUR WITH STARCH ON THE  
EVALUATION OF THE NONFAT MILK SOLIDS

Stream	Protein	Flour or Mixture Absorption	Hydration Period	Calculated Milk Absorption
	<i>g/g</i>	<i>g/g</i>	<i>min.</i>	<i>%</i>
Stone Stock	11.1	70.8	11.0	68.5
2nd Break	15.1	68.8	13.5	76.0
2nd Break plus corn starch	11.1	65.5	11.5	73.5
2nd Break plus corn starch	9.1	65.0	13.0	69.0

The addition of corn starch to the second break flour to bring the protein content to the same level as the Stone Stock gave a final evaluation of the milk solids which was better than that obtained with the Stone Stock flour.

Further dilution of the second break stream with corn starch to a protein level of 9.10% gave an evaluation of the milk solids equal to the results obtained with the Stone Stock flour.

The adverse factor in improperly processed milk solids appears to attack wheat protein from the shorter extraction streams more readily than the protein from the longer extraction streams. This effect may indicate an actual difference between proteins from various sections of the wheat kernel.

In Table I, the spring patent flours Nos. 1A to 1C, which showed a lower evaluation for the milk sample, probably contained a greater percentage of short patent streams than the other flours.

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## THE SEPARATION OF GRAIN BY PROJECTION<sup>1</sup>

ROBERT KATZ<sup>2</sup>, E. P. FARRELL<sup>3</sup>, AND MAX MILNER<sup>3</sup>

### ABSTRACT

A device is described which effects a continuous separation of internally infested wheat from sound wheat, and which separates wheat into a number of fractions of different test weight. A stream of wheat, projected into still air by rapidly moving belts, is dispersed by the combined effects of air drag and gravitation into numerous fractions which are caught in a series of hoppers. Infested kernels fall short of sound grain and are thus separated. Test weight varies progressively and characteristically with distance from the point of projection.

Recently, extensive studies have shown that the principal source of microscopic insect fragments in milled products is internal infestation (6). The removal of external insects from grain constitutes no particular problem in commercial cleaning. The milling process has been modified to reduce insect fragments in milled products about as far as presently appears economically feasible (4), and separation of internally infested grain from sound grain would be extremely desirable. At the present time there is no device in use by grain processors which effects this separation (6).

The present report deals with the development and application of an apparatus which eliminates internally infested kernels from a stream of grain and which separates grain into fractions of increasing test weight on a continuous and rapid basis.

### General Considerations

Grain cleaners in current use grade primarily by behavior in air currents and by length and width of the kernels. Insects develop within kernels of all sizes and shapes and do not affect these dimensions. An intact infested kernel may be differentiated from a sound one of the same size and shape by mass differences (10), by differences in transparency to X-rays (8), by staining of the egg plug (5, 7), and by the sounds generated by an internal insect (1).

The apparatus here described is designed to take advantage of the mass changes induced in a wheat kernel as the insect develops. A stream of kernels moving with uniform speed is projected into still

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air. Acted on by air drag and by gravitation, the grain is dispersed in such a way that infested kernels tend to fall short of sound ones of the same size and shape. The dispersed grain is collected into a series of hoppers.

Mathematical analysis of the problem is possible only when the drag is directly proportional to the velocity. At low velocities where Stokes Law applies, such analysis shows (9) that trajectories of different spheres projected from the same point with the same initial velocity are dispersed according to the parameter, mass/radius. Although most grains are not spherical, similar considerations apply. Under these conditions any separation which is effected depends not upon density (mass/volume) but upon the factor of mass/diameter. To effect a practical separation the drag forces must be greater than those prevailing under conditions where Stokes Law applies. Increased drag may be obtained by increasing the velocity of the kernel with respect to the air (2). At the velocities required for this separation, the drag force is no longer proportional to the velocity, the flow is turbulent, shape differences affect the motion, and a general mathematical analysis is not possible.

The separation principles outlined above, involving the interaction of grain with a fluid medium such as air or water, have been used for cleaning grain for centuries. Winnowing and aspiration are common examples involving the use of air as the fluid medium while the wet stoner is a familiar application involving the use of water. The present apparatus represents a carefully planned and controlled refinement of these methods.

Many of the details of the present apparatus were anticipated in a device patented almost a century ago by an inventor named J. L. Booth (3). Apparently, however, grain cleaning requirements at that time did not justify its commercial development.

### Apparatus

The grain projection device, called a grain spectrometer by the authors, is illustrated diagrammatically in Fig. 1. It consists of a hopper, *A*, with an adjustable slide-feed regulating device, two parallel belts, *B* and *C*, driven at the same speed, and a series of receiving hoppers numbered 1 to 16 into which the grain is received. The belt speed employed in these tests was approximately 1380 ft. per minute (700 cm. per second). The apparatus was adjusted to project the grain upward at an angle of approximately 25° from the horizontal. The space between the adjacent surfaces of the belts was made variable by

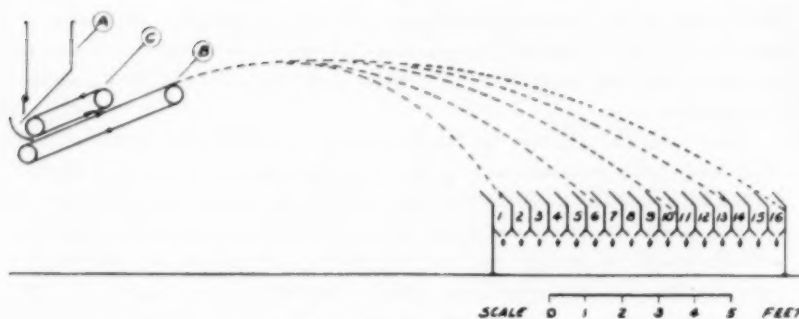


Fig. 1. The grain spectrometer.

providing adjustments for the pulley bearings of the top belt, C. As operated, this space between the two belts was approximately that of a single layer of wheat. Although the dimensions of the various components of the apparatus are not considered critical, it has been drawn to scale as shown.

### Performance

*Fractionation of Commercial Wheat on the Basis of Test Weight.* The first evaluation of the grain spectrometer involved fractionation of an ordinary sample of sound wheat. For this experiment, 5 bu. of

TABLE I  
TEST WEIGHT OF WHEAT FRACTIONS OBTAINED BY SPECTROMETRIC SEPARATION

Hopper No.	Fraction	Test Weight
	<i>oz/g</i>	<i>lb/bu</i>
Original sample	...	61.4
1	0.33	55.0
2	0.55	55.5
3	0.84	56.6
4	1.37	57.6
5	2.58	58.4
6	4.04	59.5
7	6.98	60.4
8	12.28	61.1
9	14.84	61.5
10	16.60	61.9
11	14.49	62.6
12	12.80	63.0
13	8.50	63.1
14	3.26	63.3
15	0.47	64.0
16	0.14	...

\* Quantity collected too small for test weight determination.

commercial No. 2 hard red winter wheat of 61.4-lb. test weight were used. This grain had received a preliminary cleaning by passage through a milling separator. The grain was processed in the grain spectrometer and the fractions collected in the hoppers were weighed and analyzed for test weight with a standard Boerner weight-per-bushel apparatus.

The results are shown in Table I. A regular change in test weight from 55.0 to 64.0 lb. per bushel was observed. The modal hopper was No. 10, which received 16.6% of the sample, and this fraction had a test weight of 61.9. The grain varied in appearance from thin, shriveled, and broken kernels in hoppers 1, 2, and 3 to grain of increasing plumpness in the hoppers of higher number.

To evaluate the reproducibility of the spectrometer's performance the grain which had been originally collected in hoppers numbered 6 through 13 was separately reprocessed. Specimen results are shown

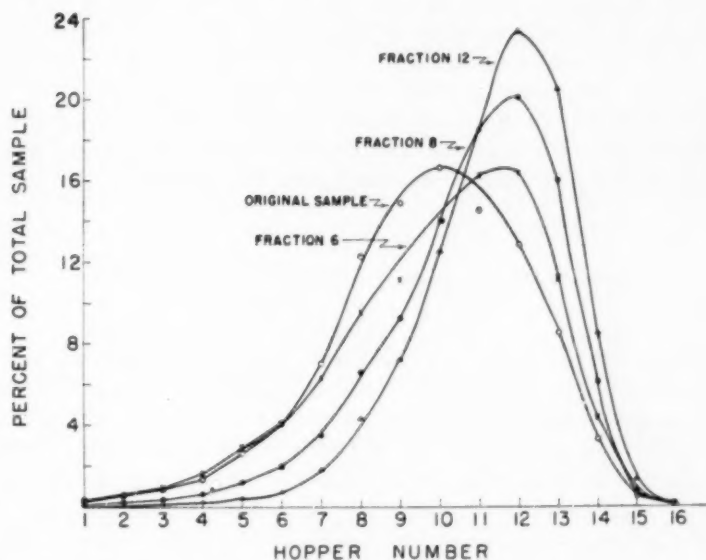


Fig. 2. Distribution of fractions from original sample and of reprocessed fractions 6, 8, and 12.

in Fig. 2. This graph shows the data for the original sample as well as for three reprocessed fractions. It is evident from these curves that the present apparatus does not separate grain with absolute precision into homogeneous and reproducible fractions. Higher hopper numbers become increasingly homogeneous in terms of the modal hopper

TABLE II  
DISTRIBUTION OF TEST WEIGHT IN FRACTIONS

Sample	Test Weight	Percent of Sample in Given Range of Test Weight		
		61 and Lower	61.1-62.5	62.6 and Higher
	<i>lb.</i>	$\frac{oz}{100}$	$\frac{oz}{100}$	$\frac{oz}{100}$
Original sample	61.4	16.7	42.2	40.7
6	59.5	51.0	44.0	5.0
7	60.4	31.2	50.7	17.8
8	61.1	23.8	53.4	23.0
9	61.5	9.9	36.9	53.0
10	61.9	4.3	40.9	53.5
11	62.6	2.3	31.0	66.6
12	63.0	... <sup>a</sup>	<14.9	85.1
13	63.1	... <sup>a</sup>	<14.3	85.7

<sup>a</sup> Quantity collected too small for test weight determination.

(No. 12) owing to the removal of low test weight fractions. Further analysis of the data resulting from the reprocessing is shown in Table II. Here the test weight of the original wheat and the test weights determined for each of the fractions 6-13 are shown. Each of these fractions was reprocessed through the spectrometer and data for test weight were broken down into three categories to show the test weight composition in the fractions. The relative amount of high test weight wheat is shown to increase markedly with bin number.

TABLE III  
SPECTROMETRIC SEPARATION OF INTERNALLY INFESTED WHEAT

Hopper No.	Fraction of Original Sample (by Weight)	Fraction Infested (by Kernel)
	$\frac{oz}{100}$	$\frac{oz}{100}$
Original sample	100.0	8.5
1	0.1	27.0
2	0.1	40.8
3	0.2	45.3
4	0.3	29.5
5	0.6	33.4
6	0.4	30.9
7	2.2	30.9
8	4.8	12.5
9	8.2	11.7
10	14.0	7.9
11	20.2	6.0
12	23.9	1.5
13	17.7	1.9
14	6.3	0.2
15	0.8	1.1
16	0.2	2.7

*Fractionation of Wheat on the Basis of Infestation.* Sixteen fractions were obtained from a sample of 16 kg. of wheat known to contain 8.5% of infested kernels. The infestation of the original wheat and the fractions was determined by radiographic inspection of 20-g. samples. The total number of kernels and the number of infested kernels shown on each radiograph were determined. The percentage distribution of fractions and percent of kernels infested appear in Table III. It is evident that a marked concentration of infested kernels occurred in the hoppers closest to the point of projection (1-7), whereas the grain falling in hoppers 12-15 was relatively free of infestation. Thus hopper 12, which received 24% of the sample, contained only 1.5% of infested kernels. Similarly, adding together all the wheat collected in hoppers 12-16, we find that 49% of the sample was reduced in infestation to approximately 1.5%, representing a six-fold purification.

Several of the radiographs from which the above data were obtained are shown (Fig. 3a-c). These supply visual evidence of the marked reduction of internal infestation achieved by one pass through the grain spectrometer.

The contents of hoppers 1-6 inclusive, which together constituted less than 2% of the total sample by weight, were about 35% infested (Table III). These fractions were discarded. The intermediate fractions were composited and reprocessed with the result that the wheat found in hoppers 12-16 was significantly cleaner than the composited as well as the original sample.

### Discussion

The ability of the grain spectrometer to separate commercial wheat into a number of test weight fractions on a continuous basis is obviously of considerable significance to the grain warehousing and milling industries. It seems probable that such apparatus could be readily integrated into the mechanical operations of mills and elevators, not only for the purpose of processing grain to provide fractions of specified test weight characteristics, but also to reclaim sound grain from commercial samples of low value.

The simplicity of the apparatus suggests its application on the farm to upgrade grain prior to marketing by improving test weight and by eliminating infested kernels. Other applications which suggest themselves but which have not yet been evaluated include separation of mixtures of grain of different species such as wheat and rye, separation of light oats from well-filled oats, separation of rodent pel-

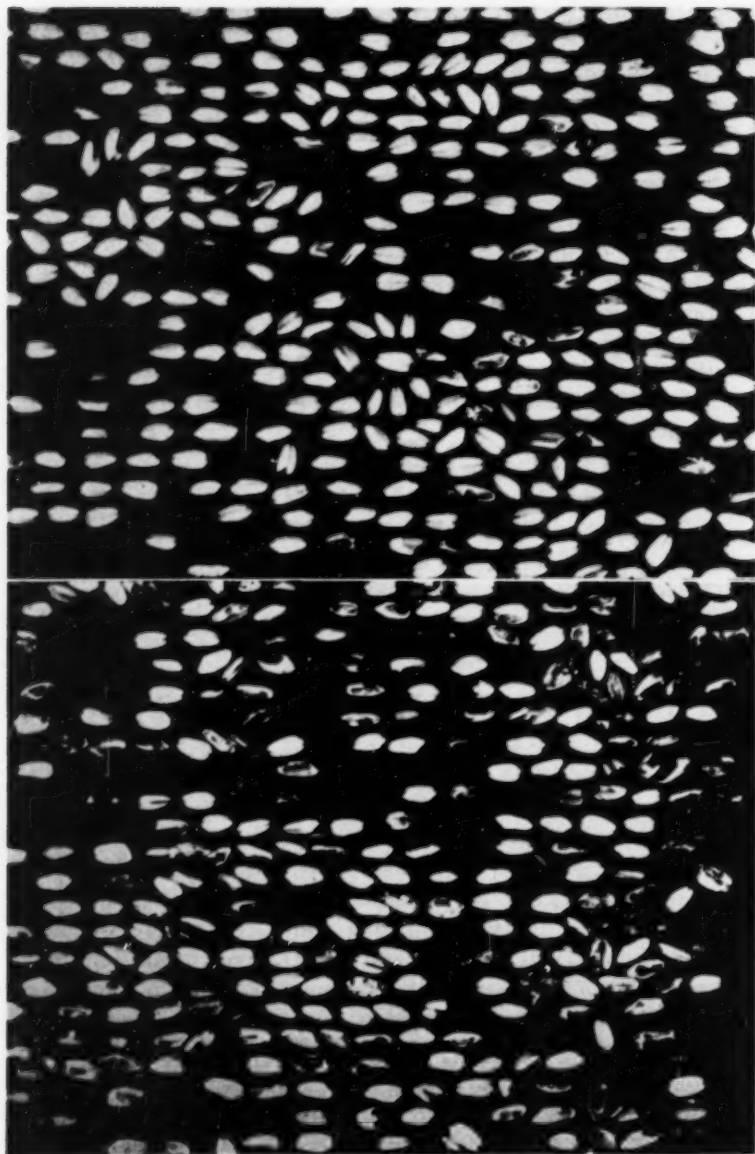


Fig. 3a. Radiographic appearance of the original sample (top) and of the fraction collected in bin 3 (bottom), showing the concentration of infestation in the bins closest to the point of projection.

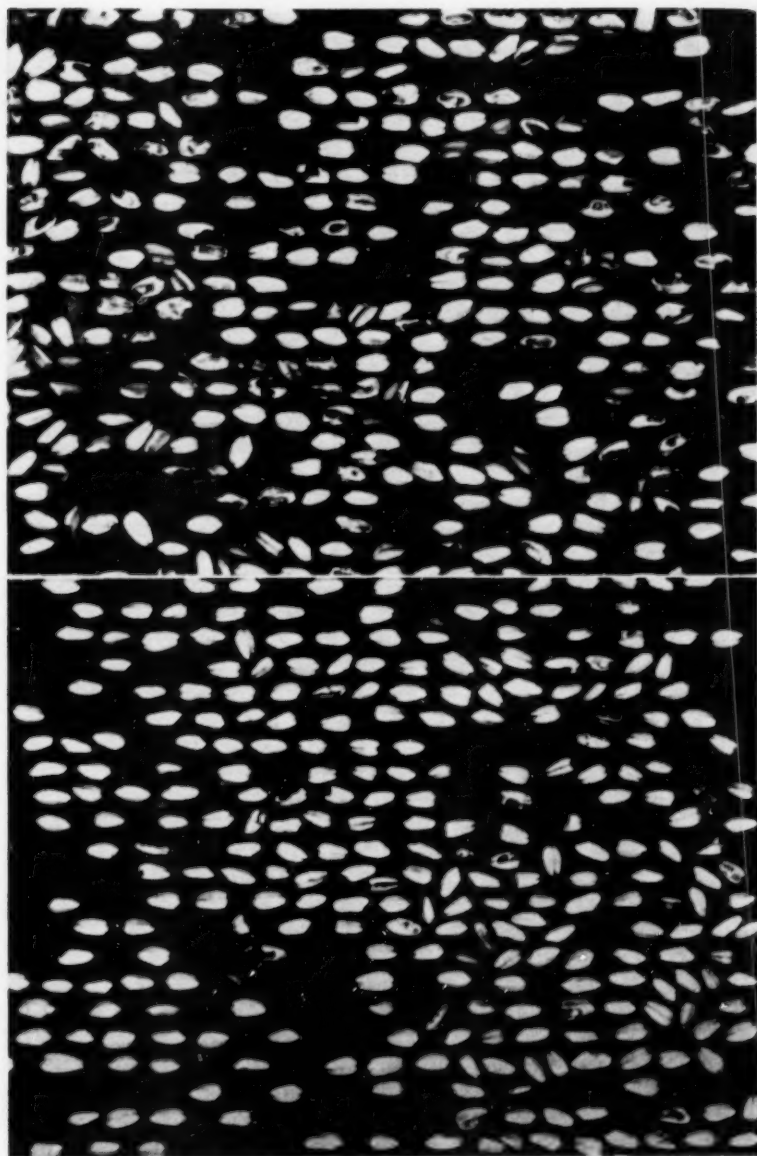


Fig. 3b. Radiographic appearance of intermediate fractions Nos. 6 (top) and 10 (bottom), showing progressive decrease in infestation with distance from the point of projection.



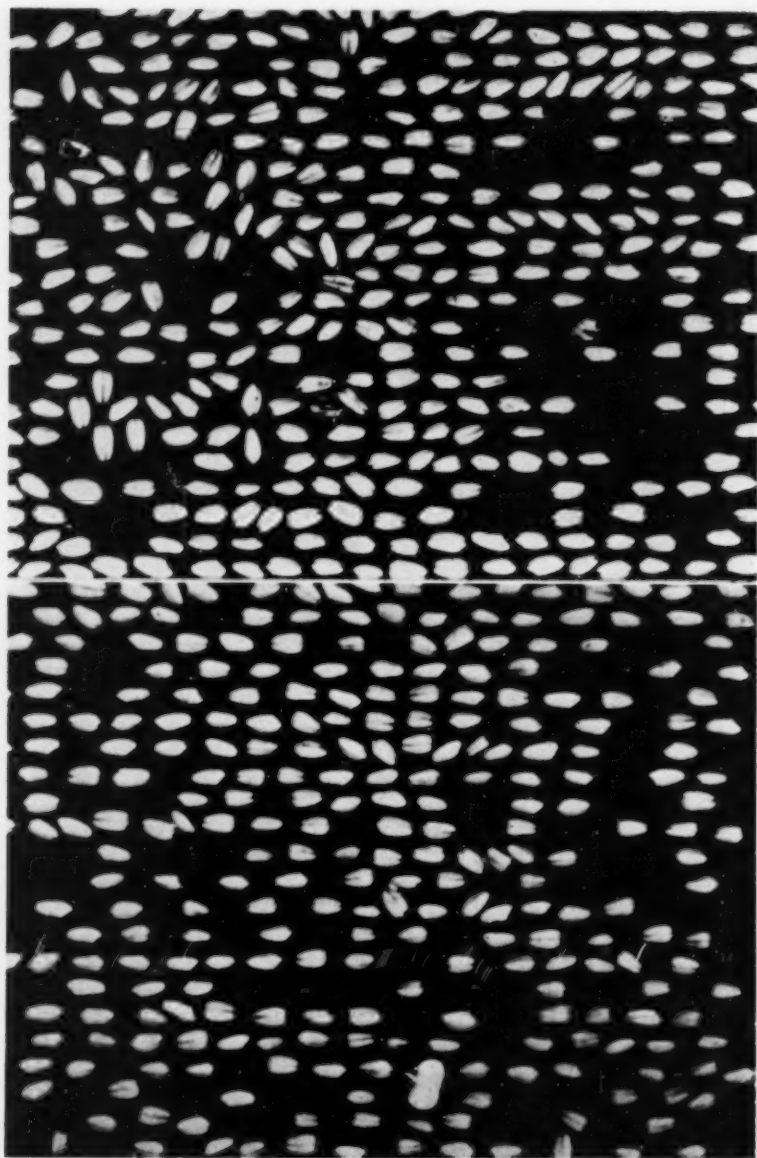


Fig. 3c. Radiographic appearance of fractions Nos. 12 (top) and 14 (bottom). Some corn kernels were segregated in the higher numbered bins. Note the relative freedom from infestation of the wheat falling in bin 14.

lets from corn and other grain, and separation of commercial seeds of all species into heavy, light, and intermediate fractions.

Differences in physical and chemical properties of the test weight fractions of a given wheat sample have been found, and the extent of these differences in terms of protein content, ash content, and technological properties of the grain will be shown in a subsequent paper.

The performance of the grain spectrometer in removing internally infested kernels suggests that a considerable amount of grain which would ordinarily be rejected for milling purposes because of excessive levels of internal infestation can be reclaimed. Thus it appears that a single pass of a lot of infested grain through such a machine would reclaim a significant proportion of the sample as sound grain. Refinements in apparatus and operating technics are now being developed which are expected to improve the resolution of the apparatus.

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## THE SEPARATION OF GRAIN BY PROJECTION. II. SYSTEMATIC DIFFERENCES IN PHYSICAL PROPERTIES AND COMPOSITION OF WHEAT FRACTIONS<sup>1</sup>

MAX MILNER,<sup>2</sup> E. P. FARRELL,<sup>2</sup> AND ROBERT KATZ<sup>3</sup>

### ABSTRACT

Analyses for test weight, 500-kernel weight, protein content, ash content, and hardness characteristics of fractions of 15 samples of hard red winter wheat, obtained by spectrometric separation, revealed high negative correlations in most cases between plumpness factors and protein and ash contents, and high positive correlations between plumpness factors and hardness, when the latter is determined with a single-stage instrument. The projection technique appears to provide a means for analyzing commercially mixed wheat for certain qualities of its components and it may be applied practically to separate either high protein fractions or high test weight fractions from ordinary commercial grain.

A preceding paper (3) has described a grain spectrometer capable of separating wheat into numerous fractions differing progressively in test weight. This development prompted a more detailed analysis of the test weight, 500-kernel weight, protein content, ash content, and hardness characteristics of fractions obtained from 15 samples of hard red winter wheat.

### Materials and Methods

The 15 samples of hard red winter wheat investigated in this study consisted of pure varieties and commercial grain grown in Kansas, Oklahoma, and Texas in the crop years 1952 and 1953. Description of these samples is given in Table I which includes their origin and test weight. Quantities of 60 lb. of each sample were processed by projection in the grain spectrometer into as many as 16 fractions. The fractions at the extreme ends of the series of receiving hoppers (see Fig. 1 of preceding paper), consisting of less than 100 g. of grain, were discarded. This produced from 12 to 14 fractions of each sample which were subjected to other tests.

Test weight was determined by means of the Boerner weight-per-bushel apparatus. A vacuum counter was used to count out 500 kernels and these were weighed to the nearest 0.01 g. Moisture, protein, and ash contents of the samples were determined as outlined in Cereal

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<sup>2</sup>Department of Flour and Feed Milling Industries.

<sup>3</sup>Department of Physics.

TABLE I  
LIST OF SAMPLES OF HARD RED WINTER WHEAT

Sample	Variety or Kind	Origin	Crop Year	Test Weight
				<i>lb.</i>
A	Commercial sample	Kansas	1952	61.6
E	Unnamed selection	Denton, Texas	1953	62.9
F	Unnamed selection	Denton, Texas	1953	61.5
H	Hd. Federation-Hybrid	Stillwater, Okla.	1953	53.5
I	Blackhull-Hd. Federation	Stillwater, Okla.	1953	54.7
J	Comanche	Stillwater, Okla.	1953	53.0
K	Unnamed selection	Manhattan, Kansas	1953	57.0
L	Cheyenne	Manhattan, Kansas	1953	57.5
M	Unnamed selection	Manhattan, Kansas	1953	56.5
N	Comanche	Manhattan, Kansas	1953	55.5
O	Ponca	Manhattan, Kansas	1953	57.1
P	Commercial sample	Abilene, Kansas	1953	55.3
Q	Commercial sample	Abilene, Kansas	1953	56.6
R	Commercial sample	Abilene, Kansas	1953	57.2
S	Commercial sample	Abilene, Kansas	1953	62.0

Laboratory Methods (2). Tests for hardness were made with the single-stage Brabender hardness tester using 200-g. samples. The curves traced on the farinograph paper were measured for maximum height, and for area by means of a planimeter.

### Results and Discussion

Results typical of those obtained with the 15 samples are shown in Table II—data for sample H listed in Table I. The hopper re-

TABLE II  
PHYSICAL AND CHEMICAL PROPERTIES OF FRACTIONS OF A SAMPLE  
OF HARD RED WINTER WHEAT

Hopper No.	Fraction of Original Sample by Weight	Test Weight	500-Kernel Weight	Protein <sup>a</sup>	Ash <sup>a</sup>	Hardness Data	
						Height	Area
	<i>%</i>	<i>lb.</i>	<i>g.</i>	<i>%</i>	<i>%</i>	<i>B.U.</i>	<i>cm.<sup>2</sup></i>
	<sup>b</sup>	53.5	8.3	17.0	2.08	380	15.5
1	0.5	°	5.2	18.9	2.26	°	°
2	0.9	46.0	5.2	17.1	2.26	°	°
3	1.6	48.0	5.4	18.5	2.27	300	13.9
4	2.7	49.0	6.4	18.8	2.28	330	14.4
5	5.0	51.0	6.3	18.5	2.25	330	13.9
6	7.4	51.8	6.8	18.3	2.19	345	14.9
7	11.3	52.7	7.3	17.8	2.16	340	14.1
8	16.4	53.5	8.1	17.5	2.14	360	14.4
9	17.0	54.2	8.5	17.3	2.13	370	15.0
10	16.5	55.1	9.1	16.6	2.10	370	15.1
11	12.4	56.1	9.7	16.0	2.09	385	15.0
12	6.3	57.3	10.4	15.2	2.01	410	16.2
13	1.8	58.0	11.3	14.4	1.97	410	15.6

<sup>a</sup> 14% Moisture basis.

<sup>b</sup> Original sample.

<sup>c</sup> Quantity too small for determination.

ceiving the largest fraction of the sample was No. 9. Considering the data for all of the 15 samples it was observed that this largest fraction could appear in any one of the hoppers numbered 9, 10, 11, or 12. There was a tendency for kernels of low test weight to fall into lower hopper numbers (closer to the point of projection) than did those of higher test weight. The fact that the modal hopper was in all cases beyond the midpoint of the 16 hoppers indicates that either a greater proportion of all the samples had mass characteristics which produced this peculiar distribution, or alternatively that the mechanical characteristics of the spectrometer were responsible for the skewed nature of the distribution. In any event the shape of the distribution curve was surprisingly uniform from sample to sample.

A common characteristic of all samples fractionated was the systematic increase in test weight and 500-kernel weight. Taking the data of Table II as typical, the fractions varied in test weight from 46 to 58 lb., a range equivalent to 22% of the test weight of the original samples. The 500-kernel weight invariably showed a greater range percentage-wise, in this case from 5.2 to 11.3 g., which is 73.5% of the original 500-kernel weight.

When plotted against hopper number the 500-kernel weights gave a more nearly linear relation than did test weights. Statistical analysis of both test weight and 500-kernel weights in relation to hopper number indicated that the variation about the linear relationship was less than 5% of the variation in test weight or kernel weight. This implies that for any given wheat whose test weight is known, the test weight of the grain falling into any hopper is predictable with a high degree of accuracy. Values for the 500-kernel weights of fractions can be predicted similarly, with precision equal to or higher than that of the test weight.

The differences in average test weight from one hopper to the next for samples H, J, P, and Q were greater than 1 lb., whereas those for varieties E, K, L, M, N, and O were less than 0.5 lb. Hence, the regression coefficient (the slope of the line relating plumpness to hopper number) is a function of the grain rather than of the machine operation.

The data for protein content are of special interest, since as indicated in Table II this sample, which contained 17.0% protein, could be fractionated into subsamples with a protein content range of 18.9 to 14.4%. It can be calculated from the data of Table II that in one pass through the spectrometer, 64% of the original sample (fractions 1 to 8 inclusive) could be recovered containing 18% protein, which is fully 1% more than that of the original grain. A number of the sam-

ples showed a similar wide range of protein in their fractions, but several showed a considerably narrower protein range.

The data for ash also are of interest, particularly from the milling viewpoint, since they indicate that within any sample the ash content, like the protein, decreases with increasing plumpness in terms of test weight, or 500-kernel weight, but to a lesser degree than does the protein content.

The height of the hardness curve as well as its area tends to increase with plumpness rather than with protein content. This is contrary to the usual assumption that protein content and hardness are directly related. These data, as will be indicated in the discussion which follows, suggest that kernel size is the major factor which determines the height and area of the hardness curves when the single-stage tester is used. The relationships apparent by inspection of Table II are indicated more quantitatively for all 15 of the samples, by means of correlation coefficients for various factors, in Table III. For purposes of interpretation, correlation values of less than 0.5 may be considered to indicate little relationship between the variables, values of 0.5 to 0.8 a moderate relationship, and values higher than 0.8 a very strong relationship.<sup>4</sup> The data of Table III show that different samples of hard red winter wheat vary considerably in intensity of correlation when only one set of factors is considered. Thus, for example, the protein-to-test-weight correlation varied from  $-0.572$  to  $-0.975$  among the 15 samples.

All the samples showed uniformly high correlations between test weight and 500-kernel weight, test weight and height of hardness curve, and 500-kernel weight and height of hardness curve. The correlations between test weight and protein content were higher than 0.8 for eight of 15 samples and higher than 0.6 for all but two of the samples, indicating that in most wheat samples of this type, a strong inverse relationship may be expected between protein content and plumpness. Essentially the same conclusion can be drawn for the relationships between plumpness and ash content. The correlations between protein and ash content were somewhat lower, although 10 of the 14 samples showed correlations between these factors of 0.6 or higher. The relations between protein content and height and area of hardness curve were rather variable from sample to sample, although in eight of 14 samples correlation values for protein and height were 0.8 or higher. High correlations between protein and curve height

<sup>4</sup>A sample correlation coefficient which exceeds the value 0.553 for these data could occur by chance only five times in 100 if the two measurements were completely unrelated, and only once in 100 times if the calculated value exceeds 0.684.



TABLE III  
CORRELATION COEFFICIENTS FOR PHYSICAL AND CHEMICAL CHARACTERISTICS OF FRACTIONS OF 15 HARD RED WINTER WHEATS<sup>a</sup>

	SAMPLE														
	A	E	F	H	I	J	K	L	M	N	O	P	Q	R	S
TW-500K	0.93	0.99	0.97	0.97	0.98	0.98	0.98	0.99	0.97	0.98	0.97	0.98	0.97	0.97	0.97
TW-Pro	-0.57	-0.59	-0.70	-0.77	-0.90	-0.87	-0.63	-0.80	-0.90	-0.10	-0.81	-0.97	-0.98	-0.94	-0.98
TW-Ash	-0.06	0.25	-0.66	-0.95	-0.95	-0.91	-0.20	-0.97	-0.51	-0.31	-0.60	-0.77	-0.96	-0.91	-0.73
TW-Ht	0.93	0.89	0.88	0.97	0.97	0.99	0.89	0.81	0.73	0.88	0.88	0.97	0.91	0.93	0.94
TW-Area	0.64	0.22	0.56	0.83	0.95	0.45	0.22	0.89	-0.17	-0.33	0.13	0.87	-0.00	0.83	0.67
500K-Pro	-0.71	-0.61	-0.79	-0.87	-0.96	-0.94	-0.60	-0.86	-0.91	-0.23	-0.91	-0.89	-0.96	-0.99	-0.98
500K-Ash	-0.03	0.29	-0.62	-0.97	-0.95	-0.97	-0.23	-0.94	-0.49	-0.24	-0.69	-0.72	-0.92	-0.89	-0.63
500K-Ht	0.93	0.91	0.93	0.98	0.95	0.99	0.89	0.79	0.78	0.90	0.81	0.98	0.97	0.90	0.97
500K-Area	0.60	0.24	0.55	0.86	0.94	0.51	0.31	0.86	-0.09	-0.33	-0.04	0.89	0.19	0.77	0.80
Pro-Ash	0.04	-0.73	0.34	0.91	0.91	0.96	-0.33	0.74	0.36	0.00	0.78	0.73	0.94	0.89	0.66
Pro-Ht	-0.09	-0.51	-0.82	-0.94	-0.94	-0.93	-0.45	-0.54	0.57	-0.68	-0.60	-0.99	-0.92	-0.90	-0.93
Pro-Area	-0.06	0.27	-0.66	-0.84	-0.95	-0.57	-0.03	-0.67	0.25	-0.04	0.21	-0.89	-0.10	-0.73	-0.69
Ash-Ht	-0.07	0.14	-0.64	-0.96	-0.96	-0.94	-0.30	-0.78	-0.60	0.14	-0.59	-0.86	-0.86	-0.70	-0.61
Ash-Area	-0.05	-0.19	-0.54	-0.86	-0.97	-0.53	0.10	-0.88	-0.52	0.41	0.08	-0.86	-0.05	-0.72	-0.21

<sup>a</sup> Abbreviations used in table headings are as follows: TW, test weight in pounds per bushel; 500K, weight in g. of 500 kernels; pro, protein content in %, 14% moisture basis; Ash, ash content in %, 14% moisture basis; Ht, maximum height of hardness curve in Brabender units; Area, area of hardness curve in cm.<sup>2</sup>.



were found more frequently than were high correlations between protein and curve areas.

The finding of a strong-inverse relationship between kernel plumpness and protein and ash content when individual samples are fractionated is in marked contrast to the virtually complete lack of correlation of these variables among different individual samples of wheat of the same class noted by Bailey and Hendel (1) as well as by Mangels and Sanderson (4). In view of the wide variation in physical and chemical properties of individual kernels within any single sample of wheat, as demonstrated in this study, the lack of correlation found by earlier workers between these factors, when *different* unfractionated samples are considered, is understandable.

The fact that the single-stage Brabender hardness tester indicated kernel plumpness to be a major factor affecting apparent hardness suggests that a study of this kind should be repeated with the two-stage tester. With this machine, all samples are ground to a uniform coarse meal prior to hardness testing.

The most significant practical implication of this study is that even grain grown on test plots is a heterogeneous mixture as regards physical properties and composition of individual kernels. The technic described makes it possible to separate commercial wheat into fractions of various protein contents, either greater or less than the protein content of the original sample. During a crop year when wheat of desirable protein content is scarce, the advantage of performing such a separation is obvious. In such a crop year kernel plumpness is usually greater than normal, and it may therefore be expected that fractions containing higher protein levels would have satisfactory plumpness characteristics. Commercial wheat which has been discounted because of low test weight can be processed also to segregate significant amounts of high test weight material, thus considerably enhancing the commercial value of a high percentage of the total.

For many years millers have sought means to separate wheat into so-called thin and plump fractions with the idea that these fractions should be processed separately in the initial or break phase of milling operations. By this means it was hoped that greater production efficiency could be attained. It would appear that the projection technic provides a practical means to accomplish this purpose. The technic might also be useful to millers for analyzing commercially mixed grain before purchase to determine the quality characteristics of the components of the mixture.

From an agronomic point of view it would seem of interest to determine the reason for the wide range of differences in wheat

characteristics, as indicated in the correlations of Table III in relation to varietal, environmental, and soil factors.

#### Acknowledgments

The authors wish to thank Mr. Henry Tucker of the Statistical Laboratory, Kansas State College, for his assistance with the statistical evaluation of the data.

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**GRAIN STORAGE STUDIES. XV.  
INFLUENCE OF MOISTURE CONTENT, COMMERCIAL  
GRADE, AND MATURITY ON THE RESPIRATION AND  
CHEMICAL DETERIORATION OF CORN<sup>1</sup>**

JON H. OLAFSON, C. M. CHRISTENSEN, AND W. F. GEDDES

**ABSTRACT**

Number 2 white dent corn respired faster at 25°C. than wheat at corresponding moistures in previous studies. It respired slowly and at a relatively constant rate over a 14-day period when the moisture content was less than 14.5%, and rapidly at an accelerating rate when the moisture content was 15.2 and 17.0%. Mold count increased at these higher moistures.

In composite samples of commercial yellow dent corn representing various grades, the fat acidities were higher, the nonreducing sugars and viability were lower, and the respiration at 30°C., particularly at moistures of 16.0% and above, was faster in the lower grades.

Prematurely harvested yellow dent corn, picked at several moisture levels as high as 46.6% and promptly air-dried at 30°C. to less than 14% moisture, respired faster than mature corn when conditioned to the same moistures. The viability of the corn was uniformly high and the fat acidity was uniformly low. Corn harvested at 46.6% moisture respired five times faster at 19.0% moisture than the most mature sample. The daily respiratory rates of the corn harvested at moisture levels of 27.9% and above increased to a maximum on the sixth day.

Unsoundness and immaturity in corn increase the storage hazard.

It is well recognized that the moisture content and soundness of corn and other grains are important factors in relation to their storage behavior. A comprehensive review of the literature on deterioration in stored corn, which was made by Semeniyuk and Gilman (10) in 1944, reveals that the heating of the grain is accompanied by mold growth and by decreases in test weight per bushel, increases in fat acidity, and other changes. Bailey (2) in 1921 found that the respiration of shelled corn was augmented by increasing the moisture content, but, in contrast with wheat, there was no sharp break in the respiration curve at a definite moisture content. Corn respired more rapidly than wheat at all moisture levels. Moreover, sprouted, heat-damaged, and cracked corn all respired more rapidly than sound corn. These early respiration studies were carried out by a static technique which involved measurements of carbon dioxide production by the grain at a controlled temperature over a fixed period of 2-4 days. Milner and Geddes (7) have pointed out that the extent of gaseous

<sup>1</sup>Manuscript received February 26, 1954. Paper No. 3128 of the Scientific Journal Series, Minnesota Agricultural Experiment Station. Cooperative investigation of the Departments of Agricultural Biochemistry and Plant Pathology. This paper represents a portion of a thesis presented to the Graduate School of the University of Minnesota in partial fulfillment of the requirements for the M.S. degree, June 1950.

exchange which occurs when grain respire in a limited supply of air may be sufficient to depress the respiration, particularly with samples of high moisture content. The relative respiratory potentials of the samples at the various moisture levels would be better reflected by means of respiration trials with continuous aeration over sufficiently long periods of time to permit mold growth in those samples having adequate moisture content.

The present paper deals with the respiration and the changes in mold count, fat acidity, and sugar content of stored corn as influenced by its moisture content, soundness, and maturity.

### Materials and Methods

A commercial sample of white dent corn (U. S. Grade No. 2) of 60% viability was employed in a preliminary study of the effect of moisture and time of storage on respiratory rate. Subsamples were conditioned to six moisture levels ranging from 13.1 to 17.8% by adding the requisite amount of distilled water and storing at 25.0°C. in a closed container for 24 hours with frequent shaking. The moisture content was determined by the two-stage, air-oven method specified by the U. S. Department of Agriculture (11).

To determine the effect of soundness on respiratory rate, composite samples of yellow dent corn were secured of grade Nos. 1 to 5 which passed through the regional offices of the Production and Marketing Administration at Cedar Rapids, Iowa; Omaha, Nebraska; Peoria, Illinois; Toledo, Ohio; and Minneapolis, Minnesota, during a 6-week period in 1947. None of the samples contained more than 14% moisture when received. Upon arrival in the laboratory, they were allowed to air dry and all samples of the same grade from the various offices were blended. Subsamples were conditioned to six moisture levels ranging from 13.5 to 19.0%, in the manner already described.

To study the effect of maturity on storage behavior, corn was harvested from a plot of hybrid yellow dent corn at six moisture levels ranging from approximately 47% to 25% in intervals of about 5%. The times of harvesting were selected after periodical moisture determinations were made on samples removed from the cob. The loss in weight of 20-30 g. dried at 130°C. for 45 minutes was determined, the sample ground, and the residual moisture determined. When the ears were picked to secure corn for the trials, several kernels were taken from each ear and bulked for moisture determination. The ears were then dried in a forced-draft oven at 30°C. until dry enough to shell. The shelled corn was then further dried at 30°C. until its

moisture content was reduced below 14% for storage, until all the samples had been assembled. For the respiration trials the samples were conditioned to 14.5, 16.5, and 19.0% moisture.

Respiratory rates for the various samples were determined at 30° C. over a period of 10 or 14 days in the apparatus described by Milner and Geddes (7). The sample weights were varied from 250 to 1500 g. and the aeration rates from 250 to 2000 ml. per 24 hours, depending upon the respiratory rates. This was done to avoid the possibility of inhibitory carbon dioxide concentrations in the interseed atmosphere and at the same time to obtain readily measurable changes in the composition of the effluent air.

Before and after the respiration trials, the samples were assayed for viability, mold count, fat acidity, reducing and nonreducing sugars. The mold counts were made by the method described by Christensen (5). Viability was determined by the Minnesota State Seed Testing Laboratory. The samples were analyzed for fat acidity by the rapid method described for corn in Cereal Laboratory Methods; reducing and nonreducing sugars were determined by the procedures for wheat flour (1).

### Results

*Effect of Moisture Content on Respiration.* The respiratory rates, at 30° C. over a 14-day period, of white dent corn of different moistures are summarized in Table I. The rates increased only slightly with time at moisture below 14.7%, but marked acceleration occurred at higher moistures. The mold count at the end of the 14-day period, which remained essentially unchanged from that of the original corn

TABLE I  
INFLUENCE OF TIME AND MOISTURE CONTENT ON THE RESPIRATORY RATE  
OF WHITE DENT CORN AT 30°C.

Day	Respiratory Rate <sup>a</sup> for Indicated Initial Moisture Content <sup>b</sup>					
	12.9	14.0	14.1	14.5	15.2	17.0
1	0.06	0.06	0.04	0.04	2.38	11.42
2	.06	.10	.07	.13	3.20	18.12
4	.05	.23	.18	.40	3.76	32.28
6	.10	.25	.23	.55	2.94	31.79
8	.10	.28	.26	.67	5.83	41.86
10	.10	.28	.32	.75	6.41	42.96
12	.07	.28	.37	.84	7.19	47.89
14	0.06	0.32	0.30	0.90	7.85	54.66

<sup>a</sup> Expressed as mg CO<sub>2</sub>/100 g dry matter/24 hours.

<sup>b</sup> The moisture contents of the samples at the end of the trial were 13.1, 13.8, 14.3, 14.7, 15.7, and 17.8% respectively.

(615,000/g) at moistures below 14.7%, had increased to 890,000/g at 15.2–15.7% moisture and to 2,000,000/g at 17.0% moisture. The viability of the corn at the end of this same period was the same as the original (60%) at moistures of 15.2% and lower; in the sample stored at 17.0% moisture the viability had decreased to 16%. These respiratory rates for corn are higher than those obtained for wheat by Milner, Christensen, and Geddes (6) at the same temperature and corresponding moistures, and are in accord with the observations of Bailey in 1921 that corn respired more rapidly than wheat at all moisture levels (2).

*Effect of Kernel Damage on Respiration and Deterioration.* Respiration trials were conducted at six moisture levels on the composite samples of the various grades of yellow corn for 10 days, and the data for the final day are given in Table II. The respiratory rates were lower than those which would be anticipated from the data in Table I. All grades respired at a low and rather uniform rate at moisture levels of 13.5, 14.5, and 15.0%, but a marked increase in rate was observed in the samples incubated at 16% and higher moistures. At the higher moistures, the lower grades of corn respired more rapidly than the higher grades. The samples of No. 4 corn at initial moisture values of 17.0 and 19.0% were exceptions to the general trend, probably because their moisture contents at the end of the trial (16.6 and 18.6% respectively) were much lower than at the beginning.

The viability, mold count, fat acidity, reducing and nonreducing sugars of the five grades of corn at the beginning and end of the respiration trials are given in Table III. In the U. S. Grain Standards for Corn, moisture is a direct grading factor and is usually the principal one which determines the grade. The moisture limits for grades

TABLE II  
RESPIRATION OF MARKET GRADES OF YELLOW DENT CORN AT DIFFERENT  
MOISTURE LEVELS ON TENTH DAY OF STORAGE AT 30°C.

Moisture Content <sup>b</sup>	Respiration Rate <sup>a</sup> for Grade No.				
	1	2	3	4	5
13.5	0.03	0.08	0.18	0.11	0.11
14.5	0.08	0.17	0.18	0.23	0.29
15.0	0.12	0.28	0.27	0.33	0.55
16.0	0.60	2.00	1.93	2.09	4.95
17.0	5.91	25.23	23.07	19.73	33.22
19.0	69.03	94.06	108.01	99.40	115.36

<sup>a</sup> Expressed as mg CO<sub>2</sub>/100 g dry matter/24 hours.

<sup>b</sup> Initial moisture content of corn when placed in respirometer. The values at the end of the trial were all within 0.1% of the original figures except No. 4 corn at 17.0 and 19.0% moisture, which contained 16.6 and 18.6% moisture, respectively.

TABLE III

EFFECT OF STORING VARIOUS MARKET GRADES OF YELLOW DENT CORN AT DIFFERENT MOISTURE CONTENTS FOR 10 DAYS AT 30°C. ON MOLD COUNT, VIABILITY, FAT ACIDITY, AND SUGAR CONTENT

Initial Moisture	U. S. Grade No.				
	1	2	3	4	5
	Mold count, in Thousands/g				
Original	11	525	11	220	3,000
13.5	5	355	260	420	3,000
14.5	15	61	168	75	850
15.0	80	17	15	113	1,000
16.0	40	400	360	113	1,750
17.0	6	15,000	1,000	1,180	750
19.0	480	800	950	10,000	8,500
	Viability				
Original	65	46	44	42	30
13.5	66	50	42	44	30
14.5	70	50	55	48	25
15.0	64	50	38	48	25
16.0	60	48	32	37	23
17.0	60	32	27	37	20
19.0	25	15	15	8	5
	Fat acidity, mg KOH/100 g corn (d.m. basis)				
Original	22	38	43	55	77
13.5	24	37	45	66	71
14.5	32	38	56	71	76
15.0	25	36	65	53	74
16.0	29	39	56	52	72
17.0	25	48	59	59	82
19.0	48	68	79	79	120
	Nonreducing sugars, as mg sucrose/10 g corn (d.m. basis)				
Original	143	121	118	113	102
13.5	134	134	124	118	100
14.5	135	110	124	106	93
15.0	135	118	136	103	92
16.0	141	116	138	108	81
17.0	125	102	121	109	72
19.0	106	50	105	109	41
	Reducing sugars, as mg maltose/10 g corn (d.m. basis)				
Original	41	41	44	46	58
13.5	43	55	61	49	48
14.5	44	56	56	48	47
15.0	48	50	60	45	44
16.0	45	52	46	59	50
17.0	41	52	63	54	52
19.0	60	54	36	50	44



1 to 5 are 14.0, 15.5, 17.5, 20.0, and 23.0%, respectively. It is not surprising therefore that, in general, the fat acidity of the composite samples increased with a decrease in grade, whereas the viability and nonreducing sugars decreased. Only the samples stored at 19% moisture showed marked changes in any of these properties as a result of incubation at 30° C. for the 10-day period. The mold counts at 14.5 and 15.0% moisture were lower than those for the corresponding grades at 13.5% moisture. This may have been due to the germination of some of the spores on the samples at 13.5% moisture soon after the conditioning water was added, followed by their death when the moisture diffused into the kernel. *Penicillium* was the predominant species in the original seeds of all grades and also in the majority of the samples after incubation. In grades 1 and 5 stored at 17% moisture, and grades 2 and 3 at 19% moisture, *Aspergillus flavus* comprised 50% to 100% of the spores found at the end of the storage trial.

*Effect of Maturity.* Respiratory rates for the corn harvested at six

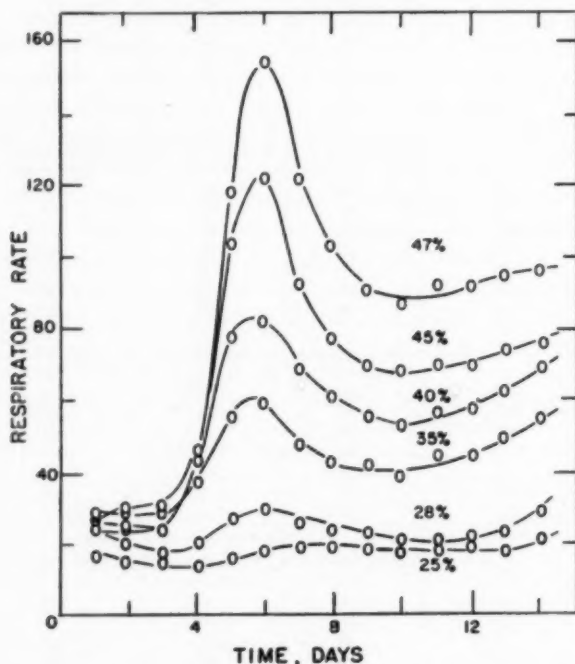


Fig. 1. The effect of moisture content at harvest on the daily respiratory rates of corn at 30° C. Respiratory rate is expressed as mg. carbon dioxide produced per 100 g. dry matter per 24 hours. The moisture contents designating the various curves are only approximate; the actual values are given in Table IV.

stages of maturity (46.6 to 24.9% moisture) were determined at 14.5, 16.5, and 19.0% moisture for 14 days at 30° C. The daily respiration values for the trials at 19.0% moisture are plotted in Fig. 1. The total carbon dioxide produced in 14 days at the three moisture levels is given in Table IV, together with the viability, mold count, and fat acidity of the samples before and after the trials.

The respiratory rates of corn harvested at 27.9 to 46.6% moisture reached a maximum on the sixth day followed by a sharp decrease to a minimum on the tenth day; thereafter a gradual increase in respiration occurred until the end of the trials on the fourteenth day. The more immature the corn at harvest, the higher was the respiratory rate; in the respiration trials at 19.0% moisture, the sample harvested at 46.6% moisture exhibited a fivefold greater respiration than the most mature sample (24.9% moisture). For each stage of maturity, sig-

TABLE IV  
INFLUENCE OF MATURITY ON THE RESPIRATION, VIABILITY, MOLD COUNT,  
AND FAT ACIDITY OF CORN UPON INCUBATION AT DIFFERENT  
MOISTURES FOR 14 DAYS AT 30° C.

Sampling Date	Moisture at Harvest	Incubation Moisture <sup>a</sup>	Respiration <sup>b</sup>	Viability	Mold Count <sup>c</sup>	Fat Acidity <sup>d</sup>
	%	%		%		
Aug. 30	46.6	original	...	95	9.5	26
		14.5	3	92	...	25
		16.5	116	90	262	27
		19.0	1160	48	4,800	112
Sept. 2	44.7	original	...	97	12.5	21
		14.5	4	98	12	22
		16.5	208	94	1,000	25
		19.0	942	58	21,000	62
Sept. 7	40.1	original	...	98	1.3	21
		14.5	3	98	0.7	18
		16.5	70	96	59	22
		19.0	773	71	3,100	39
Sept. 13	34.6	original	...	98	16.2	16
		14.5	2	97	3	16
		16.5	55	96	19.7	16
		19.0	601	80	3,000	34
Sept. 23	27.9	original	...	97	...	20
		14.5	2	95	...	21
		16.5	25	96	...	20
		19.0	326	83	...	26
Sept. 29	24.9	original	...	98	...	20
		14.5	2	98	...	20
		16.5	25	97	...	20
		19.0	243	89	...	22

<sup>a</sup> "Original" indicates corn before respiration trials.

<sup>b</sup> Mg CO<sub>2</sub>/100 g dry matter for 14 days.

<sup>c</sup> Expressed as thousands of mold colonies per g. of corn.

<sup>d</sup> Mg KOH/100 g dry matter.

nificant increases in mold count occurred when the respiration trials were conducted at 16.5 and 19.0% moisture; at the latter moisture, the viability of the samples decreased and the fat acidity increased.

### Discussion

The results of these experiments are in agreement with observations in the literature that the storage properties of grain are influenced by such factors as the moisture content, the degree of maturity at harvest, and the soundness of the grain when placed in storage (2-4, 8-10). Immature corn or grain which has been subjected to harvesting and handling conditions that result in varying degrees of unsoundness will respire more rapidly than mature sound grain when stored at the same moisture contents. The hazards involved in storing immature and unsound grain are therefore much greater than those involved with corn of high quality.

### Acknowledgments

The authors are indebted to the Grain Branch, Production and Marketing Administration (now designated as Agricultural Marketing Service), for its cooperation in providing composite samples of various commercial grades of corn, and to the Department of Agronomy, Minnesota Agricultural Experiment Station, for growing the corn used in the maturity studies.

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# A NOTE ON THE SPECIFIC HEAT OF RICE, OATS, AND THEIR PRODUCTS<sup>1</sup>

G. A. HASWELL<sup>2</sup>

A study of the relationship between the specific heat and the moisture content of Bersée wheat and other grains has been made by R. W. Disney (1) in this laboratory. Since the technical literature contains no information on this relationship for rice and oats, the experiments reported here were carried out with the modified Bunsen ice calorimeter and method described by Disney. The results are recorded in Table I.

TABLE I  
EFFECT OF MOISTURE CONTENT ON THE SPECIFIC HEATS OF RICE,  
OATS, AND THEIR PRODUCTS

Sample No.	Moisture Content (Wet Wt. Basis)	Specific Heat, Mean	Number of Replicates	Standard Error
	<i>%</i>	<i>Cals/g/°C</i>		
<i>Rough Rice, (Italian Origin)</i>				
1	17.0	.450	12	.0023
2	13.5	.402	15	.0046
3	10.2	.378	12	.0033
<i>Shelled Rice, (Italian Origin)</i>				
4	17.6	.447	12	.0039
5	14.5	.418	12	.0025
6	9.8	.376	12	.0049
<i>Fully Finished Rice, (Italian Origin)</i>				
7	17.4	.440	12	.0034
8	14.6	.411	12	.0029
9	10.8	.380	12	.0045
<i>Sun II Oats</i>				
10	17.8	.445	12	.0043
11	14.8	.419	12	.0038
12	11.7	.397	12	.0033
<i>Sun IX Groats</i>				
13	17.6	.466	16	.0047
14	11.8	.397	12	.0040

Samples Nos. 2, 5, 8, 12, and 14 were not conditioned before the specific heat determinations were made. Samples Nos. 1, 4, 7, 10, 11, and 13 were conditioned to their respective moisture contents by

<sup>1</sup> Manuscript received March 18, 1934.

<sup>2</sup> Dept. of Scientific and Industrial Research, Pest Infestation Laboratory, London Road, Slough, Bucks, England.

adding distilled water and shaking. Thereafter, they were allowed to equilibrate for at least one week. Samples Nos. 3, 6, and 9 were dried in a vacuum over calcium chloride until they reached their respective moisture contents.

Moisture contents were determined by drying triplicate ground samples in a ventilated oven at 113°C. for 4 hours.

Straight line regressions were calculated (Table II), since it was evident that most of the data are best represented in this way. Disney's data for wheat are also clearly linear over the moisture range covered in the present data.

TABLE II  
REGRESSION EQUATIONS FOR THE RELATION BETWEEN SPECIFIC HEATS  
AND MOISTURE CONTENT

Grain	Regression Equation*	Apparent Specific Heat of Water (cals/g/°C)
Rough rice	$C = 0.0107M + 0.265$	1.335
Shelled rice	$C = 0.0091M + 0.287$	1.197
Fully finished rice	$C = 0.009 M + 0.282$	1.182
Oats	$C = 0.0078M + 0.305$	1.085
Groats	$C = 0.0119M + 0.257$	1.447

\*C = Apparent specific heat (cals/g/°C); M = moisture content, %.

The results for rough rice were less well-fitted by a straight line than are those for the other grains tested, a fact which may be related to the extreme thickness of the husk.

The apparent specific heat of water was calculated from each of these regressions and, with the exception of oats, the values lay close to those obtained by Disney for Bersée wheat in the same range of moisture content.

The lines for shelled and fully finished rice were at different levels but parallel; this may be due to a difference in their specific heats at zero moisture content, related to the specific heat of the rice bran.

Oat groats gave a higher regression coefficient than oats; this may be related to the higher fat content (8.1%) of groats in comparison with oats (4.5%) and rice (0.4%–2.0%) (2).

#### Acknowledgments

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## BOOK REVIEWS

**Foodstuffs, their Plasticity, Fluidity, and Consistency.** Edited by G. W. Scott Blair, xii + 264 pp. North-Holland Publishing Co., Amsterdam, and Interscience Publishers, Inc., New York. 1953.

This volume contains chapters on the rheology of miscellaneous products and a general chapter on psychorheology of foodstuffs. Cereal chemists will be particularly interested in Chapter II on Cereals, by D. H. Greup and H. M. R. Hintzer (The Netherlands), and to a lesser extent in Chapter I on Starch, by J. Hofstee and A. H. A. De Willigen (The Netherlands), and Chapter VIII by R. Harper (England), as well as in selected details scattered throughout the remainder of the book.

The chapter on the rheology of dough and bread (44 pp.) is an excellent summary of the subject. Fundamental principles of dough rheology are discussed by reviewing mainly the pioneer work of Scott Blair and his associates. Another section of the chapter is devoted to the rheology of breadmaking. It begins with a consideration of the mixing process, goes on to discuss the changes resulting from fermentation, and baking, and concludes with the rheology of bread crumb in relation to staling. The final section of the chapter describes the common dough-testing instruments: farinograph and mixograph; extensograph, extensometer and alveograph; and amylograph.

The chapter on psychorheology of foodstuffs reviews the relationship between objective measurements and subjective assessment. A short section (5 pp.) is devoted to an analysis of bakers' judgment of dough and bread. The chapter also includes a discussion of a statistical technic called "Factor Analysis" and its application to the study of rheology of foodstuffs.

The authors and editor are to be commended on the clarity of presentation and on timely emphasis on fundamental rheology. This small volume will be a welcome addition to the literature on dough rheology which is just beginning to emerge as a coherent field of study.

I. HLYNKA,  
Grain Research Laboratory,  
Board of Grain Commissioners for Canada

**Storage of Cereal Grains and Their Products.** Edited by J. A. Anderson and A. W. Alcock, 600 pp., American Association of Cereal Chemists, Monograph Series, Volume II. 1954. Price, \$11.00.

To the undersigned has been given the opportunity of contributing a review of this, the second monograph issued by the American Association of Cereal Chemists. Few can know better than the writer the increasing difficulty of attempting to deal, in one comprehensive book, with the many aspects of what is still known as cereal chemistry—a name already really out of date but nevertheless, because of usage, convenient. While there is still a demand for books that broadly cover the whole subject, the need for more detailed specialized knowledge on sections of the subject is a very real one. This was shown by the success of the first monograph of the Association, *Enzymes and Their Role in Wheat Technology*, and it also accounts for the impatience with which many of us have awaited this second and most-difficult-to-write monograph. Its production, with no less than fourteen authors involved, has been a time-consuming affair but few will doubt, when they receive the book, that it has been worth waiting for.

The subjects (covered in separate chapters) and the authors are: I, Moisture and its measurement, by I. Hlynka and A. D. Robinson; II, Chemical, physical and nutritive changes during storage, by Lawrence Zeleny; III, Microflora, by G. Semeniuk; IV, Respiration and heating, by Max Milner and W. F. Geddes; V, Insects, by R. T. Cotton; VI, Rodents, by Donald A. Spencer; VII, Country storage of grain, by H. J. Barre; VIII, Terminal elevator storage, by J. E. Bailey; IX, Drying of grain, by W. V. Hukill; X, Flour storage in bulk, by R. E. Hamilton, C. J.



Lynde, and R. K. Larmour; XI, Packaging and storage of cereal products, by C. A. Southwick, Jr.

This book will obviously attract as much attention outside North America as it will in that region. It has been plentifully reviewed there, but many in Europe and elsewhere may be interested in how the book strikes a non-American.

The present reviewer has little doubt that it will be most welcomed, since it is probably the most comprehensive book so far published on an important and fascinating subject. Further, it has to deal with many matters that are debatable and yet are of great importance economically.

Having said that, one must also state that the book is obviously written by North Americans for North Americans and that the main background is the experience gained in the United States and Canada. The considerable experience on many of the problems which exist in Europe is often neglected, even though on some aspects Europe may have acquired more knowledge than America. This criticism, of course, applies only to certain practical aspects of grain and flour storage and its handling and not to the scientific background of the subject. In a book contributed by so many authors there is always unevenness, depending not only on the individual's style of writing but on the general approach to the subject and the breadth of view and knowledge of the contributor. This is clearly shown in various chapters where the bibliography is almost entirely restricted to the work of Americans, often to the neglect of important work done elsewhere; in other chapters a wider view is taken and proper appreciation of the work of others is given. One feels that the use of essentially American references is not a matter of prejudice by the particular contributor but may reflect lack of knowledge of what has been done elsewhere.

It is impossible to deal with all the chapters individually. Some of the early chapters deal with the scientific problems involved; then follow chapters dealing with insect infestation (and what greater authority is there than R. T. Cotton?) and rodents; and finally come a series of chapters dealing with certain practical aspects of storage as practiced in North America, such as "Country storage of grain" and "Terminal elevator storage," these latter naturally relating to American practice.

To European readers Chapter IX on the "Drying of grain" may be of particular interest. This is a subject which constantly has to be faced in Europe, where often the grain is harvested containing 18%-20% moisture and, for storage, this has to be reduced to 12%-13%. The first portion of this interesting chapter deals with theoretical considerations and therefore there is common ground all the world over; but the final portion, dealing with actual American practice, may seem somewhat remote to the European miller who has special knowledge of the problem when the grain is really damp.

European readers with their somewhat longer experience of flour storage in bulk will read with particular interest Chapter X. Useful as this is, there appears no reference to flat-bottomed dischargers of the Redler type nor of the "fluidization" of flour and similar powders by means of a comparatively small amount of air pressure. It may well be that this system, which is in practice in Europe, will become a matter of fundamental importance.

No volume, particularly on such a complicated and changing subject, can hope to be perfect or even to be free from minor defects and criticism. The cereal world will, without any doubt, welcome this monumental work and large numbers of people will remain grateful to the Association for its great endeavor. There can be few workers and students in this field who will not treasure this volume in their libraries.

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## Cereal Chemistry

### EDITORIAL POLICY

**Cereal Chemistry** publishes scientific papers dealing with raw materials, processes, or products of the cereal industries, or with analytical procedures, technological tests, or fundamental research, related thereto. Papers must be based on original investigations, not previously described elsewhere, which make a definite contribution to existing knowledge.

**Cereal Chemistry** gives preference to suitable papers presented at the Annual Meeting of the American Association of Cereal Chemists, or submitted directly by members of the Association. When space permits, papers are accepted from other scientists throughout the world. The papers must be written in English and must be clear, concise, and styled for **Cereal Chemistry**.

Manuscripts for publication should be sent to the Editor in Chief. Advertising rates may be secured from and subscriptions placed with the Managing Editor, University Farm, St. Paul 1, Minnesota. Subscription rates, \$11.00 per year. Foreign postage, 50 cents extra. Single copies, \$2.50; foreign, \$2.60. Back issues, \$3.00.

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### SUGGESTIONS TO AUTHORS

**General.** Authors will find the last volume of *Cereal Chemistry* a useful guide to acceptable arrangements and styling of papers. "On Writing Scientific Papers for Cereal Chemistry" (*Trans. Am. Assoc. Cereal Chem.* 6:1-22. 1948) amplifies the following notes.

Authors should submit two copies of the manuscript, typed double spaced with wide margins on 8½ by 11 inch white paper, and all original drawings or photographs for figures. If possible, one set of photographs of figures should also be submitted. Originals can then be held to prevent damage, and the photographs can be sent to reviewers.

**Titles and Footnotes.** Titles should be specific, but should be kept short by deleting unnecessary words. The title footnote shows "Manuscript received . . ." and the name and address of the author's institution. Author footnotes, showing position and connections, are desirable although not obligatory.

**Abstract.** A concise abstract of about 200 words follows title and authors. It should state the principal results and conclusions, and should contain, largely by inference, adequate information on the scope and design of the investigation.

**Literature.** In general, only recent papers need be listed, and these can often be cited more advantageously throughout the text than in the introduction. Long introductory reviews should be avoided, especially when a recent review in another paper or in a monograph can be cited instead.

References are arranged and numbered in alphabetical order of authors' names and show author, title, journal, volume, first and last pages, and year. The list is given at the end of the paper. Reference numbers must invariably be cited in the text, but authors' names and year may be cited also. Abbreviations for the names of journals follow the list given in *Chemical Abstracts* 45: VII-CCLV (1951).

**Tables.** Data should be arranged to facilitate the comparisons readers must make. Tables should be kept small by breaking up large ones if this is feasible. Only about eight columns of tabular matter can be printed across the page. Authors should omit all unessential data such as laboratory numbers, columns of data that show no significant variation, and any data not discussed in the text. A text reference can frequently be substituted for columns containing only a few data. The number of significant figures should be minimized. Box and side headings should be kept short by abbreviating freely; unorthodox abbreviations may be explained in footnotes, but unnecessary footnotes should be avoided. Leader tables without a number, main heading, or ruled lines are often useful for small groups of data.

Tables should be typed on separate pages at the end of the manuscript, and their position should be indicated to the printer by typing "(TABLE I)" in the appropriate place between lines of the text. (Figures are treated in the same way.)

**Figures.** If possible, all line drawings should be made by a competent draftsman. Traditional layouts should be followed: the horizontal axis should be used for the independent variable; curves should be drawn heaviest, axes or frame intermediate, and the grid lines lightest; and experimental points should be shown. Labels are preferable to legends. Authors should avoid identification in cut-lines to be printed below the figure, especially if symbols are used that cannot readily be set in type.

All drawings should be made about two to three times eventual reduced size with India ink on white paper, tracing linen, or blue-lined graph paper; with any other color, the unsightly mass of small grid lines is reproduced in the cut. Lettering should be done with a guide using India ink; and letters should be 1/16 to 1/8 inch high after reduction.

All original figures should be submitted with one set of photographic reproductions for reviewers, and each item should be identified by lightly writing number, author, and title on the back. Cut-lines (legends) should be typed on a separate sheet at the end of the manuscript. "Preparation of Illustrations and Tables" (*Trans. Am. Assoc. Cereal Chem.* 3: 69-104, 1945) amplifies these notes.

**Text.** Clarity and conciseness are the prime essentials of a good scientific style. Proper grouping of related information and thoughts within paragraphs, selection of logical sequences for paragraphs and for sentences within paragraphs, and a skillful use of headings and topic sentences are the greatest aids to clarity. Clear phrasing is simplified by writing short sentences, using direct statements and active verbs, and preferring the concrete to the abstract, the specific to the general, and the definite to the vague. Trite circumlocutions and useless modifiers are the main causes of verbosity; they should be removed by repeated editing of drafts.

**Editorial Style.** A.A.C.C. publications are edited in accordance with *A Manual of Style*, University of Chicago Press, and *Webster's Dictionary*. A few points which authors often treat wrongly are listed below:

Use names, not formulas, for text references to chemical compounds. Use plural verbs with quantities (6.9 g. were). Figures are used before unit abbreviations (3 ml.), and % rather than "per cent" is used following figures. All units are abbreviated and followed by periods, except units of time, which are spelled out. Repeat the degree sign ( $5^{\circ}$ - $10^{\circ}$ C.). Place 0 before the decimal point for correlation coefficients ( $r = 0.95$ ). Use \* to mark statistics that exceed the 5% level and \*\* for those that exceed the 1% level; footnotes explaining this convention are no longer required. Type fractions on one line if possible, e.g.,  $A/(B + C)$ . Use lower case for farinograph, mixogram, etc., unless used with a proper name, i.e., Brabender Farinograph. When in doubt about a point that occurs frequently, consult the Style Manual or the Dictionary.

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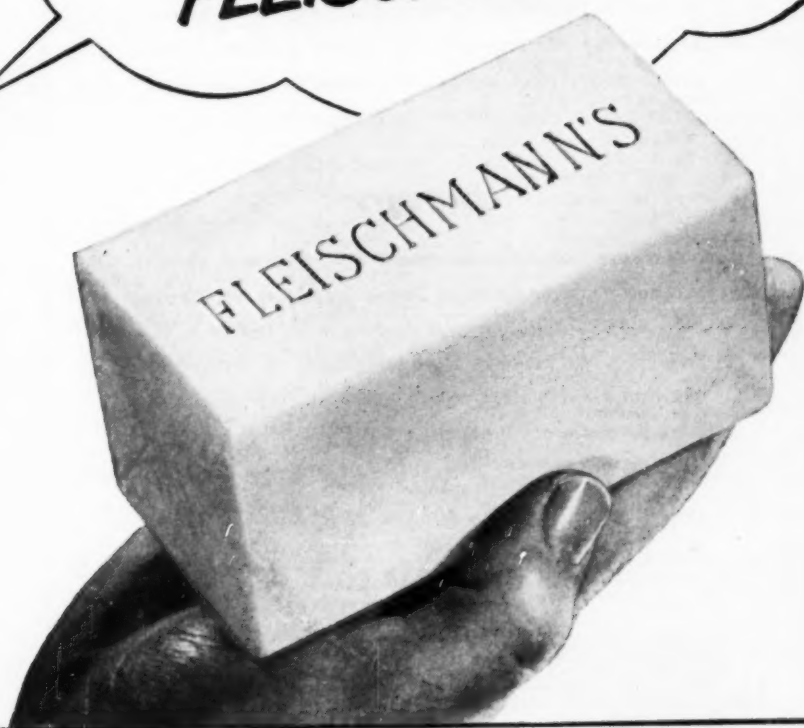
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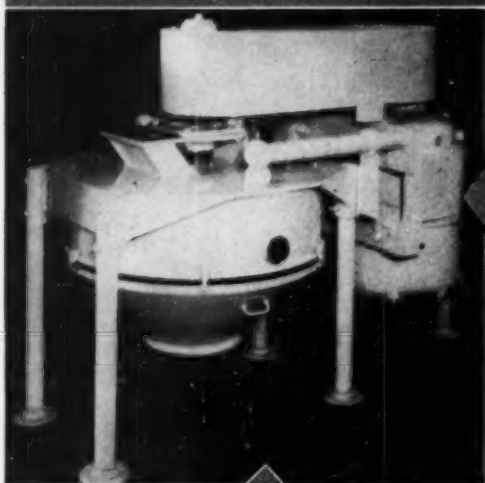


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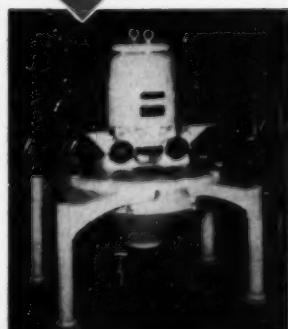


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